REMARKS

Claims 21-24, 28-38, 40-42, 46, 48-54, 56, 57, 63-66, and 68-75 are currently pending in the application. Claims 1-20, 25-27, 39, 43-45, 47, 55, 58-62, and 67 have been cancelled. No claims are amended in this response.

Rejections under 35 U.S.C. § 103

Claims 21-24, 28-38, 40-42, 46, 48-54, 56, 57, 63-66, and 68-75 remain rejected as unpatentable over Muller and Andersson. The rejections are traversed.

I. Initial Burden Is on the Examiner to Establish a Prima Facie Case of Obviousness

The Examiner has maintained the rejections of the claims over US 5,858,410 ("Muller) and US 5,739,152 ("Anderson") stating that "there is no evidence of record that the addition of a surfactant would alter the function of the claimed composition and would materially change the nature of the claimed composition." Applicants respectfully traverse.

The test of obviousness turns on whether the Examiner has provided factual evidence for the rejections. In *In re Warner*, the Federal Circuit held:

> The Patent Office has the initial duty of supplying the factual basis for its rejection. It may not, because it may doubt that the invention is patentable, resort to speculation, unfounded assumptions or hindsight reconstruction to supply deficiencies in its factual basis.[1]

The Final Office Action fails the *In re Warner* test of requiring the factual basis for the rejection.[2] In concluding that "there is no evidence of record" the Examiner overlooked the supporting arguments presented in the response filed on April 21, 2011. Applicants, therefore, invite the Examiner to offer the factual basis for the assertion.

Because the rejection fails in the fundamental tenet of an obviousness rejection, Applicants submit that the Examiner has expressed doubt only by resorting to speculation and

[1] *See In re Warner*, 379 F.2d 1011, 1016 (CCPA 1967).
[2] *Id.*

unfounded assumptions to supply the deficiencies in the factual basis presented by the Applicants.[3]

In the interest of furthering prosecution only, Applicants further provide *Lawrence et al.* (2000) -- published a year after filing the current application – which states that Tween (polyoxyethylene surfactant) based microemulsion are not suitable for autoclaving because of the surfactant's characteristic phase inversion temperature (PIT).[4] Lawrence suggests substituting autoclaving with filtration in the presence of a surfactant:

> Although it is considered that the polyoxyethylene surfactants [Tween] are the most sensitive, other non-ionic surfactants . . . are also sensitive to changes in temperature.
>
> The presence of a PIT can cause problems for the exploitation of microemulsions stabilised by nonionic surfactants as drug delivery systems. **This is a particular problem where . . . autoclaving**, the preferred means of sterilisation, **is likely to destabilise the microemulsions**. However, sterilization by filtration remains an option for low viscosity droplet-containing microemulsions.[5]

Because of the characteristic PIT of Tween 80, the viscosity of the emulsion would increase during autoclaving. Muller, thus, included Tween 80 to increase viscosity of particles prepared by autoclaving.[6] Applicants, therefore, submit that it is well known in the art that emulsions comprising surfactants are not suitable for autoclaving when low viscosity is desired. Solely based on this common knowledge, one of ordinary skill would have avoided using surfactant in the composition of the current invention.

Anderson does not remedy the deficiency in Muller. The Final rejection thus failed in finding a *prima facie* obviousness. Applicants, therefore, request that the rejections be withdrawn.

---

[3] *See In re Warner*, 379 F.2d 1011, 1016 (CCPA 1967).
[4] *See Lawrence et al.*, "Microemulsion-based media as novel drug delivery systems." *Adv. Drug Delivery Rev.* 45: 89-121 (2000).
[5] *Id.* at 95. [Emphasis added.]
[6] *See* Response to Non-Final Office Action filed on April 21, 2011, at page 11, ¶ 4.

II. All Words of a Claim Must be Considered in an Obviousness Rejection

To support an obviousness rejection, MPEP § 2143.03 requires "all words of a claim to be considered" and MPEP § 2141.02 requires consideration of the "[claimed] invention and prior art as a whole." Further, the Board of Patent Appeal and Interferences confirmed that a proper, post-KSR obviousness determination still requires that the Office make "a searching comparison of the claimed invention – including all its limitations – with the teaching of the prior art."[7] In sum, it remains well-settled law that an obviousness rejection requires at least a suggestion of all of the claim elements. Because the current obviousness rejection ignores claim element "particle size of up to 3 μm, with not more than 3000 particles of a size of 10 μm or greater and not more than 300 particles of a size of 25 μm or greater" of claims 21 and 38, Applicants submit that the rejection is improper.

Anderson does not remedy the deficiency in Muller. The Final rejection thus failed in finding a *prima facie* obviousness. Applicants, therefore, request that the rejections be withdrawn.

Claims 23, 24, 29-33, and 37 depend on claim 21 and incorporates each and every element of the claim. Claims 40-42, 46-54, 56-57, 63-66, and 68-75 depend on claim 38 and incorporates each and every element of the claim. The dependent claims are non-obvious when the claims they depend on are non-obvious.[8]

For at least the above reasons, Applicant respectfully submits that the Office Action fails to set forth a *prima case* of obviousness. Applicant, therefore, requests the Examiner to withdraw the rejections and allow the claims.

---

[7] *In re Wada and Murphy*, Appeal 2007-3733 (citing *In re Ochiai*, 71 F.3d 1565, 1572 (Fed. Cir. 1995), *and CFMT v. Yieldup Intern. Corp.*, 349 F.3d 1333, 1342 (Fed. Cir. 2003)).

[8] *See* MPEP § 2143.03 ("If an independent claim is nonobvious under 35 U.S.C. 103, then any claim depending therefrom is nonobvious." (citing *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988).)

Applicants submit that the application is in condition for allowance and request an action for the same. A petition for an extension of time accompanies this response. Please charge any additional fees due, or credit any overpayment of same, to Deposit Account 50-0311, Attorney Reference No.: 28069-503001US.

Respectfully submitted,

David E. Johnson, Reg. No. 41,874
Attorney for Applicant
c/o MINTZ LEVIN
Tel: 617-542-6000
Fax: 617-542-2241
Customer No. 30623

Date: August 30, 2011

5345996v.3

ELSEVIER

Advanced Drug Delivery Reviews 45 (2000) 89–121

advanced drug delivery reviews

www.elsevier.com/locate/drugdeliv

Microemulsion-based media as novel drug delivery systems

M. Jayne Lawrence[a,*], Gareth D. Rees[b,*]

[a]*Department of Pharmacy, King's College London, Franklin Wilkins Building, 150 Stamford Street, London SE1 9NN, UK*
[b]*SmithKline Beecham R&D, St. George's Avenue, Weybridge, Surrey KT13 0DE, UK*

Abstract

Microemulsions are clear, stable, isotropic mixtures of oil, water and surfactant, frequently in combination with a cosurfactant. These systems are currently of interest to the pharmaceutical scientist because of their considerable potential to act as drug delivery vehicles by incorporating a wide range of drug molecules. In order to appreciate the potential of microemulsions as delivery vehicles, this review gives an overview of the formation and phase behaviour and characterization of microemulsions. The use of microemulsions and closely related microemulsion-based systems as drug delivery vehicles is reviewed, with particular emphasis being placed on recent developments and future directions.

Contents

*Co-corresponding authors. Tel.: +44-0207-848-4808; fax: +44-0207-848-4800 (M.J. Lawrence). Tel.: +44-1932-822179; fax: +44-1932-822120 (G.D. Rees).
E-mail addresses: jayne.lawrence@kcl.ac.uk (M.J. Lawrence), gareth.d.rees@sb.com (G.D. Rees).

0169-409X/00/$ – see front matter
PII: S0169-409X(00)00103-4

1. Introduction

The microemulsion concept was introduced as early as the 1940s by Hoar and Schulman who generated a clear single-phase solution by titrating a milky emulsion with hexanol [1]. Schulman and coworkers (1959) subsequently coined the term microemulsion [2], and it has since been defined and indeed redefined on many occasions. For the purposes of this review, however, the microemulsion definition provided by Danielsson and Lindman in 1981 will be used as the point of reference [3]. Microemulsions are thus defined as 'a system of water, oil and amphiphile which is a single optically isotropic and thermodynamically stable liquid solution.'

In practice, the key difference between emulsions and microemulsions are that the former, whilst they may exhibit excellent kinetic stability, are fundamentally thermodynamically unstable and will eventually phase separate [4]. Another important difference concerns their appearance; emulsions are cloudy while microemulsions are clear or translucent. In addition, there are distinct differences in their method of preparation, since emulsions require a large input of energy while microemulsions do not. The latter point has obvious implications when considering the relative cost of commercial production of the two types of system.

It is also useful to note that under the definition given, self-microemulsifying drug delivery systems (SMEEDS) are not microemulsions, although they may be considered to be a closely related system. A SMEDD typically comprises a mixture of surfactant, oil and drug (known as the concentrate) which when introduced into the body is rapidly dispersed to form droplets of approximately the same size range as those observed in microemulsion systems. Once dispersed such systems would be expected to behave in vivo much the same way as oil-in-water (o/w) microemulsions.

The above broad definition does not require a microemulsion to contain any microstructure, in other words it includes systems that are co-solvents, that is, systems in which the constituent components are molecularly dispersed. Most researchers in the field agree however that for a microemulsion to be formed it is important that the system contains some definite microstructure, in other words there is a definite boundary between the oil and water phases at which the surfactant is located. In order to gain an understanding of the reasons for microemulsion formation, it is first useful to consider the properties of amphiphiles, such as surfactants, in solution.

Conventional surfactant molecules comprise a polar head group region and an apolar tail region, the latter having the larger molecular volume particularly in the case of ionic surfactants. On dispersal in water, surfactants self-associate into a variety of equilibrium phases, the nature of which stems directly from the interplay of the various inter and intermolecular forces as well as entropy considerations. Surfactants also self-associate in non-aqueous solvents, particularly apolar liquids such as alkanes. In this case the orientation of the surfactant molecules are reversed compared to those adopted in aqueous solution. This reorientation serves to optimise the solvation requirements of the surfactant and minimises the free energy of the system overall. When surfactants are incorporated into immiscible mixtures of oil and water, the surfactant molecules can locate at the oil/water interface which is thermodynamically very favourable. A number of phases can result which may be structured on the microscopic or macroscopic scale, one example of a phase structured on the microscopic scale is an optically isotropic microemulsion phase. The schematic given in Fig. 1 gives an indication of a few of the wide variety of possible self-association structures that surfactants can form in the presence of water, oil or combinations of all three. Although outside the scope of this review many of the structures shown in Fig. 1, as well as some of those not shown, have potential for use as drug delivery systems.

Fig. 2 shows schematic representations of the three types of microemulsions which are most likely

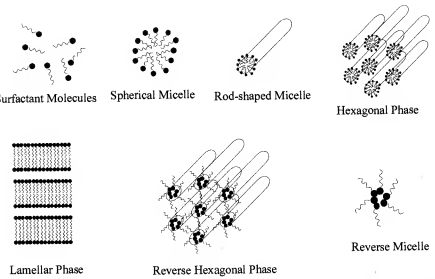

Fig. 1. Schematic representation of the most commonly encountered self-association structures in water, oil or a combination thereof.

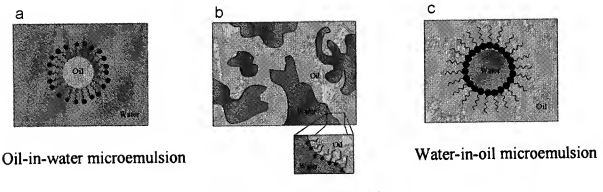

Fig. 2. Schematic representation of the three most commonly encountered microemulsion microstructures: (a) oil-in-water, (b) bicontinuous, and (c) water-in-oil microemulsion.

to be formed depending on composition. It can be seen while the three structures shown are quite different, in each there is an interfacial surfactant monolayer separating the oil and water domains. Note that while the oil-in-water (o/w) and water-in-oil (w/o) droplets are represented in Fig. 2 as spheres, they may be asymmetric in shape, frequently adopting the shape of a prolate ellipsoid. The presence of o/w microemulsion droplets is likely to be a feature in microemulsions where the volume fraction of oil is low. Conversely, w/o droplets are likely when the volume fraction of water is low, and in systems where the amounts of water and oil are similar, a bicontinuous microemulsion may result. In the latter case, both oil and water exist as a continuous phase in the presence of a continuously fluctuating surfactant-stabilised interface with a net curvature of zero.

The relationship between micelles and o/w microemulsion droplets as well as between reverse

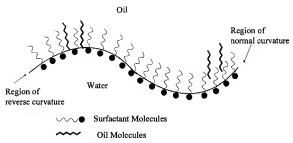

Fig. 3. Penetration of oil molecules between the hydrophobic chains of the interfacial surfactant monolayer in a bicontinuous microemulsion. Note the greater extent of oil penetration when the film curves towards water (i.e. in a region of reverse curvature). Modified from [235].

micelles and w/o microemulsion droplets has been debated on a number of occasions. Clearly there is a transition through the series (reverse) micelle, swollen micelle and microemulsion droplet but by definition micelles and reverse micelles are not microemulsions. Distinguishing between swollen micelles and microemulsion droplets is largely a semantic exercise, but it is recognised that as the ratio of dispersed phase to surfactant increases, the physicochemical properties approach those of the pure solvent. It should be noted that while the definition used in the present study does not differentiate between a swollen micelle and a microemulsion, other researchers in the field do however make this distinction.

Depending upon the nature of the oil, in particular its size relative to the hydrophobic chain of the surfactant, the oil may penetrate to varying extents into the surfactant tails of the interfacial monolayer. This is shown schematically in Fig. 3 for a bicontinuous microemulsion; a similar effect has been proposed to occur in both o/w and w/o microemulsions.

2. Overview of microemulsion formation and phase behaviour

2.1. Theories of microemulsion formation

Historically, three approaches have been used to explain microemulsion formation and stability. These are: (i) interfacial or mixed film theories [2,5]: (ii) solubilisation theories [6–8]: and (iii) thermodynamic treatments [9–11]. An in depth discussion of these theories are beyond the scope of this review but has been addressed by others [12]. However, an admittedly simplified thermodynamic rationalisation is presented below. The free energy of microemulsion formation can be considered to depend on the extent to which surfactant lowers the surface tension of the oil–water interface and the change in entropy of the system such that,

$$\Delta G_f = \gamma \Delta A - T \Delta S$$

where ΔG_f is the free energy of formation, γ is the surface tension of the oil–water interface, ΔA is the change in interfacial area on microemulsification, ΔS is the change in entropy of the system which is effectively the dispersion entropy, and T is the temperature. It should be noted that when a microemulsion is formed the change in ΔA is very large due to the large number of very small droplets formed. Originally workers proposed that in order for a microemulsion to be formed a (transient) negative value of γ was required, it is now recognised that while value of γ is positive at all times, it is very small (of the order of fractions of mN/m), and is offset by the entropic component. The dominant favourable entropic contribution is the very large dispersion entropy arising from the mixing of one phase in the other in the form of large numbers of small droplets. However, there are also expected to be favourable entropic contributions arising from other dynamic processes such as surfactant diffusion in the interfacial layer and monomer-micelle surfactant exchange. Thus a negative free energy of formation is achieved when large reductions in surface tension are accompanied by significant favourable entropic change. In such cases, microemulsification is spontaneous and the resulting dispersion is thermodynamically stable.

Qualitatively we know that several factors determine whether a w/o or o/w system is formed. Intuitively, we would summise that the most likely microemulsion would be that in which the phase with the smaller volume fraction forms the droplets, and indeed this is very often although by no means exclusively the case. By their very nature, o/w microemulsion droplets generally have a larger effective interaction volume than w/o droplets. In the

case of ionic surfactants this is attributable to the presence of an electrical double layer at the surface of the o/w droplet which introduces a strong repulsive term. For o/w microemulsions stabilised by a non-surfactant, although there is hydration shell associated with the polar headgroups, the predominant repulsive factor can be attributed to steric interactions. Additionally, it is pertinent to note that it is easier to arrange surfactant at an interface with high curvature, i.e., small droplets, if the surfactant tails extend outwards into a continuous oil phase. This is also entropically more favourable as the hydrocarbon tails have more directional freedom. As a result, interfacial tension tends to be lower for a w/o microemulsion than for an o/w microemulsion, thereby making their preparation a more facile process. It should also be remembered however, that while microemulsions are thermodynamically stable there may be kinetic barriers to their formation. As a consequence, the order of component addition may impact on the ease of preparation, and in some cases mechanical agitation or the input of heat will assist more rapid microemulsification.

2.2. Phase behaviour

The relationship between the phase behaviour of a mixture and its composition can be captured with the aid of a phase diagram. Compositional variables can also be studied as a function of temperature and pressure, although with the exception of microemulsions prepared using supercritical or near critical solvents [13,14], or with liquefied chlorofluorocarbon [15] and HFA propellants [16], the large majority of systems are studied under conditions of ambient pressure. The phase behaviour of simple microemulsion systems comprising oil, water and surfactant can be studied with the aid of ternary phase diagram in which each corner of the diagram represents 100% of that particular component. More commonly, however, and almost always in the case of microemulsions in pharmaceutical applications, the microemulsion will contain additional components such as a cosurfactant and/or drug. The cosurfactant is also amphiphilic with an affinity for both the oil and aqueous phases and partitions to an appreciable extent into the surfactant interfacial monolayer present at the oil–water interface. The cosurfactant need not necessarily be capable of forming association structures in its own right. A wide variety of molecules can function as cosurfactants including non-ionic surfactants [17,18], alcohols [19,20], alkanoic acids, alkanediols and alkyl amines [21]. Surprisingly few studies have examined the effect of drug on phase behaviour, this is despite the fact that a large number of drug molecules are themselves surface active [22] and as such would be expected to influence phase behaviour. Fig. 4 shows the effect of the presence of drug (either fenoprofen or fenoprofen sodium) on the phase behaviour of reverse phospholipid aggregates.

In the case where four or more components are investigated, pseudo-ternary phase diagrams are used where a corner will typically represent a binary mixture of two components such as surfactant/cosurfactant, water/drug or oil/drug. The number of different phases present for a particular mixture can be visually assessed. Microstructural features can also be investigated with the aid of a wide variety of techniques, which are discussed later in this review. A highly schematic (pseudo) ternary phase diagram illustrating these features is presented in Fig. 5. It should be noted that not every combination of components produce microemulsions over the whole range of possible compositions, in some instances the extent of microemulsion formation may be very limited.

Constructing phase diagrams is time consuming, particularly when the aim is to accurately delineate a phase boundary, as the time taken for the system to equilibrate can be greatly increased as the phase boundary is approached. Heat and sonication are often used, particularly with systems containing nonionic surfactants, to speed up the process. The procedure most often employed is to prepare a series of (pseudo) binary compositions and titrate with the third component, evaluating the mixture after each addition. Care must be taken to ensure not only that the temperature is precisely and accurately controlled, but also that observations are not made on metastable systems. Clearly, however, time constraints impose a physical limit on the length of time systems can be left to equilibrate and consequently the elimination of metastable states can be difficult to ensure in practice, although centrifugation can be useful to speed up any separation. References to

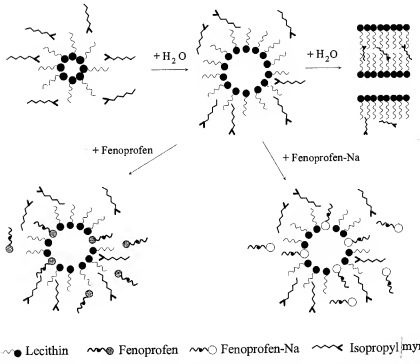

Fig. 4. Schematic representation of the effect of drug on the association of phospholipid in an isopropylmyristate (IPM) system. Top, from left to right: phospholipid molecules in IPM in the absence of water and drug form ellipsoidal reverse micelles, increasing the amount of water present causes these aggregates to transform first into rod-like micelles and then finally lamellar liquid crystals. Bottom, from left to right: drug solubilisation either in its free acid or sodium salt form causes a change in shape of the colloidal aggregates — with fenoprofen acid rod like micelles transform into more spherical ones, with fenoprofen salt rod-like micelles transform into extremely long rods. Modified from [222].

rapid screening procedures have appeared in the literature [23].

Outside the microemulsion region, particularly for compositions close to the oil–water binary axis, there is insufficient surfactant to facilitate the formation of a single microemulsion phase. In this case multiple phases may exist, the complexity of which increases with the number of components in the mixture. Within this region, and indeed other multiphase regions of the ternary phase diagram, microemulsions can exist in equilibrium with excess water or oil phases. These multiphase systems can be conveniently described using the Winsor classification [24]. In the Winsor classification, the one phase microemulsions that are generally explored as drug delivery systems are known as Winsor IV systems.

Transitions between the various phases mapped out in these phase diagrams can be driven by the further addition of one of the components, addition of a new component such as drug or electrolyte, or by changing the temperature. Transitions from w/o to o/w microemulsions may occur via a number of different structural states including bicontinuous, lamellar and also multiphase systems. Microemulsions stabilised by non-ionic surfactants, especially those based on polyoxyethylene, are very susceptible to temperature because a decrease in surfactant solubility occurs with increasing temperature, and as a result systems stabilised by non-ionic surfactants or mixtures thereof often have characteristic phase inversion temperatures (PITs), with the PIT of the microemulsion varying with a range of experimental factors including the amount and nature of the oil present and the nature of the surfactant(s) present. This latter case is well illustrated in Fig. 6 which shows the effect on PIT of increasing the amount of

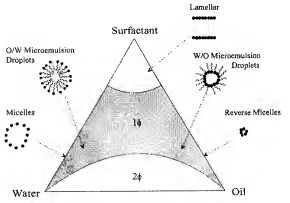

Fig. 5. A hypothetical pseudo-ternary phase diagram of an oil/surfactant/water system with emphasis on microemulsion and emulsion phases. Within the phase diagram, existence fields are shown where conventional micelles, reverse micelles or water-in-oil (w/o) microemulsions and oil-in-water microemulsions are formed along with the bicontinuous microemulsions. At very high surfactant concentrations two phase systems are observed.

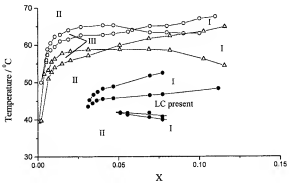

Fig. 6. Effect of temperature and weight fraction of surfactant mixture on the phase behaviour of a water/$C_{12}E_4$/sucrose dilaurate/heptane system. The weight fraction of the lipophilic sucrose dilaurate in the surfactant mixture are 0.5 (closed circles), 0.7 (open triangle) and 0.8 (open circle). The water/heptane ratio is 50/50 (w/w). I, II and III indicate one-, two-, and three phase regions, with the one phase region being the microemulsion region. Note that as the proportion of the lipophilic sucrose dilaurate increases, the I phase microemulsion region is formed at higher temperatures. Redrawn from [131].

the lipophilic sugar surfactant present in a mixed $C_{12}E_8$:sucrose dilaurate system. It should be noted that the presence of electrolyte or in some cases drug, especially if lipophilic in nature, can act to lower the PIT, illustrating the importance of determining microemulsion phase behaviour in the presence of drug. Although it is considered that the polyoxyethylene surfactants are the most sensitive, other non-ionic surfactants such as the alkylamine-*N*-oxides and the sugar surfactants are also sensitive to changes in temperature. In contrast microemulsions stabilised by ionic surfactants have little or no sensitivity to temperature.

The presence of a PIT can cause problems for the exploitation of microemulsions stabilised by non-ionic surfactants as drug delivery systems. This is a particular problem where formulations are intended for intravenous administration as autoclaving, the preferred means of sterilisation, is likely to destabilise the microemulsion. However, sterilisation by filtration remains an option for low viscosity droplet-containing microemulsions. In order to avoid any complications due to the presence of a PIT, the intended temperature of use should be 30 K below the PIT. Although it is possible to envisage circumstances whereby the presence of a phase transition in use can be used to derive a benefit, for example to release drug at a pre-determined site in the body.

2.3. The role of surfactant

The single-phase microemulsion systems of interest in this review are classified as Winsor IV. The surfactants used to stabilise such systems may be: (i) non-ionic, (ii) zwitterionic, (iii) cationic, or (iv) anionic surfactants. Combinations of these, particularly ionic and non-ionic, can be very effective at increasing the extent of the microemulsion region. Examples of non-ionics include polyoxyethylene surfactants such as Brij 35 ($C_{12}E_{23}$) or a sugar esters such as sorbitan monooleate (Span 80). Phospholipids are a notable example of Zwitterionic surfactants and exhibit excellent biocompatibility. Lecithin preparations from a variety of sources including soybean and egg are available commercially and contain diacylphosphatidylcholine as its major constituent [19,21,25,26]. Quaternary ammonium alkyl salts form one of the best known classes of cationic surfactants, with hexadecyltrimethylammonium bromide (CTAB) [27–29], and the twin-tailed surfactant didodcecylammonium bromide (DDAB) amongst the most well known [30–32]. The most widely studied anionic surfactant is probably sodium bis-2-ethylhexylsulphosuccinate (AOT)

which is twin-tailed and is a particularly effective stabiliser of w/o microemulsions [18,30,33–37].

Attempts have been made to rationalise surfactant behaviour in terms of the hydrophile–lipophile balance (HLB) [38], as well as the critical packing parameter (CPP) [39,40]. Both approaches are fairly empirical but can be a useful guide to surfactant selection. The HLB takes into account the relative contribution of hydrophilic and hydrophobic fragments of the surfactant molecule. It is generally accepted that low HLB (3–6) surfactants are favoured for the formation of w/o microemulsions whereas surfactants with high HLBs (8–18) are preferred for the formation of o/w microemulsion systems. Ionic surfactants such as sodium dodecyl sulphate which have HLBs greater than 20, often require the presence of a cosurfactant to reduce their effective HLB to a value within the range required for microemulsion formation.

In contrast, the CPP relates the ability of surfactants to form particular aggregates to the geometry of the molecule itself. The CPP can be calculated using the following equation:

$$CPP = v/a.l$$

where v is the partial molar volume of the hydrophobic portion of the surfactant, a is the optimal head group area and l is the length of the surfactant tail. The latter parameter is often expressed as l_c, that is the critical length of the hydrophobic chain, generally assumed to be 70–80% of its fully extended length. The CPP is a measure of the preferred geometry adopted by the surfactant, and as a consequence is predictive of the type of aggregate that is likely to form. The effect of changing CPP is illustrated in Fig. 7 but put simply, cone-shaped surfactants will pack at curved interfaces whereas surfactants whose geometry can be represented by truncated cones or rectangular blocks prefer to form worm-like micelles or lamellar structures [41]. Of

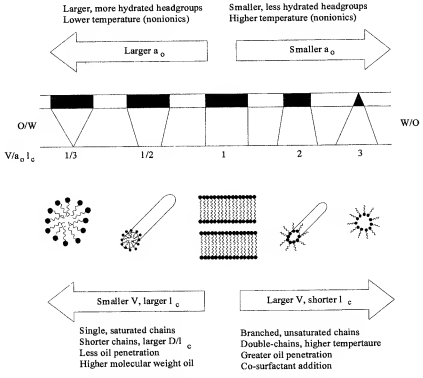

Fig. 7. Effect of molecular moieties and solution conditions on the CPP of a surfactant and the resulting range of possible surfactant aggregates in water or aqueous solution. Redrawn from [236].

course, changes in microemulsion composition will modify the microenvironment of the surfactant, which will lead to changes in the apparent CPP of the surfactant. For example in a microemulsion system, penetration of small oil molecules between the hydrocarbon tails would be expected to increase the effective surfactant hydrophobe volume, whereas large molecular volume oils would not be expected to exert much effect on the CPP [42]. Similarly, increases in ionic strength would be expected to result in a decrease in the effective head group area of ionic surfactants as the double layer shrinks and screening of the head groups allows closer approach. The presence of hydrophilic molecules such as glycerol and sorbitol in the aqueous phase will also influence optimal head group area by altering the solubility of the head group in the aqueous phase. Because of these effects, water-soluble hydrophilic materials have been used as to aid microemulsion formation as can be seen in Fig. 8. However, whilst such materials may be referred to as cosurfactants, this description is misleading, as they are not amphiphilic in their own right. As with traditional cosurfactants their presence can lead to destruction of the microemulsion upon dilution. Finally, the effect of temperature on these parameters is especially pertinent for non-ionics such as polyoxyethylene alkyl ethers as the polyoxyethylene (PEO) group is dehydrated with increasing temperature. This has the effect of altering substantially the CPP and in extremis is manifested by phase separation or phase inversion.

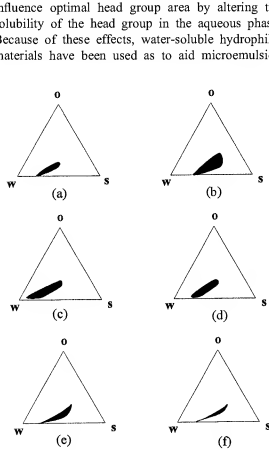

Fig. 8. Partial phase diagrams of the system isopropyl myristate (O), polysorbate 40 and sorbitol (S) and water (W) showing areas of existence of o/w microemulsions at 37°C for polysorbate/sorbitol mass ratios of a, 1/1; b, 1/1.5; c, 1/2 d, 1/2.5; e, 1/3; and f, 1/3.5. Redrawn from [86].

In most cases, single-chain surfactants alone are unable to reduce the oil/water interfacial tension sufficiently to enable a microemulsion to form, a point made in a number of pertinent microemulsions reviews [43–48]. Medium chain length alcohols which are commonly added as cosurfactants, have the effect of further reducing the interfacial tension, whilst increasing the fluidity of the interface thereby increasing the entropy of the system [44,45,48]. Medium chain length alcohols also increase the mobility of the hydrocarbon tail and also allow greater penetration of the oil into this region. Furthermore, any alcohol present may also influence the solubility properties of the aqueous and oily phases due to its partitioning between these phases. It has also been suggested that some oils, for example the ethyl esters of fatty acids, also act as 'cosurfactants' by penetrating the hydrophobic chain region of the surfactant monolayer [49]. All of the aforementioned mechanisms are considered to facilitate microemulsion formation. In the case of microemulsions stabilised by ionic surfactants, the addition of alkanols also serves to reduce repulsive interactions between the charged head groups.

A number of double chain surfactants such as AOT and DDAB are able to form microemulsions without the aid of cosurfactants [18,30–37,50,51]. These surfactants are characterised by having small head groups in comparison to their hydrocarbon tails. Phosphatidylcholine or lecithin is also a twin-tailed surfactant, but in this case it is generally necessary to include a cosurfactant in order to disrupt the lamellar structures which characterise its biological behaviour. Thus medium chain alcohols have been successfully used as cosurfactants for the formation of

lecithin-based microemulsions [19,26]. Interestingly w/o microemulsions have been prepared using short diacyl chain lecithins and small molecular volume oils where it is possible that the small molecular volume oils penetrate the hydrophobic chain region thereby facilitating microemulsion formation [52].

3. Microemulsion characterisation

In contrast to their ease of preparation, it is a far from trivial matter to characterise the microstructure of a microemulsion, yet such knowledge is essential for their successful commercial exploitation. For example, it has been shown that the rate of release of sodium salicylate from a lecithin-based microemulsions, is dependent upon their microstructure [53].

Microemulsions have been evaluated using a wide range of different techniques over the years, but a complementarity of methods is generally required in order to fully characterise these systems. At the macroscopic level viscosity, conductivity and dielectric methods provide useful information [29,35,36,54–56]. Viscosity measurements for example can indicate the presence of rod-like or worm-like reverse micelles [55,56], and conductivity measurements provide a means of determining whether a microemulsion is oil-continuous or water-continuous, as well as providing a means of monitoring percolation or phase inversion phenomena [37,55,57]. Dielectric measurements are a powerful means of probing both the structural and dynamic features of microemulsion systems [37,58,59].

The isotropic nature of microemulsions and their optical clarity makes their study by spectroscopic techniques straightforward, particularly in comparison to conventional macroemulsions. Pulsed field gradient NMR for example has been used extensively to measure self-diffusion coefficients of the various components and yields information on the mobility and microenvironment [25,28,32,38,56,60–68]. Scattering methods have also been invaluable in elucidating microemulsion structure and methods employed include dynamic and static light scattering [16,28,52,55,62,69–74], small-angle neutron scattering (SANS) [35,60,62,75–79] and small-angle X-ray scattering (SAXS) [30,60–62,76,79,80]. Indeed the value of scattering methods is exemplified by the work of Tabony who identified the presence of a cubic phase in the bicontinuous microemulsion region [77,78]. These techniques have also been extremely useful in the development of microemulsion models such as the cubic random cell (CRC) [76,81] and disordered open connected (DOC) models [30,79]. Neutron scattering methods using contrast variation have been used to probe the nature of the oil penetration into the interfacial surfactant monolayer of the microemulsion [82]. Freeze-fracture electron microscopy has also been used to study microemulsion structure, however extremely rapid cooling of the sample is required in order to maintain structure and minimise the possibility of artifacts [75,83–85].

A potentially serious limitation with some of these methods of analysis lies in the requirement to dilute the microemulsion systems in order to eliminate particle–particle interactions. This is because dilution can drive a phase transition or a molecular reorganisation, and is therefore a particular problem for techniques such as viscometry, NMR self-diffusion measurements and those relying on scattering. It is therefore often necessary to work with systems containing a relatively high dispersed phase concentration and to account for interparticulate interactions by use of a model. On the one hand this provides an opportunity to extract useful information regarding particle–particle interactions, but on the downside it to makes the structural characterisation of concentrated systems extremely problematic.

In spite of the abovementioned complications, much of the work reported in the pharmaceutical literature has been conducted using concentrated microemulsion systems. For the most part where particle sizes obtained using photon correlation spectroscopy, the measurements quoted remain uncorrected, not least because such corrections are far from trivial. Only a few studies have attempted to correct for these interactions [86,87], yet without such corrections, these data should only be used to establish the presence of microemulsion structure. Whilst neglecting correction factors, some workers have attempted to correlate apparent droplet sizes obtained in concentrated systems with oral bioavailability. Unsurprisingly, this approach has met with very limited success.

4. Microemulsion-based systems in drug delivery

Microemulsions have generated considerable interest over the years as potential drug delivery systems [43–48,88,89]. Advantages associated with microemulsions include their thermodynamic stability, optical clarity and ease of preparation. The existence of microdomains of different polarity within the same single-phase solution enables both water-soluble and oil-soluble materials to be solubilised, and at the same time if this is so desired. Furthermore it is also possible to incorporate amphiphilic drugs into the microemulsion, sometimes even leading to an increase in the extent of existence of the microemulsion region. It should be noted that solubilisate partitions between the microemulsion droplet and continuous phase and that while there may be a preferred site of solubilisation within the microemulsion droplet, solubilisate may be located at one of a number of sites. For example the likely preferred sites of incorporation of a lipophilic, water-insoluble drug into an o/w microemulsion are the disperse oil phase and/or hydrophobic tail region of the surfactant molecule, while a water-soluble material would be most likely to be incorporated into the disperse aqueous phase of a water-in-oil droplet. In some instances the viscosity of the microemulsion may be tailored for a given application through formulation changes, or in some instances through the incorporation of specific gelling agents such as Carbopol [90] or gelatin [91] as shown in Fig. 9.

Table 1 illustrates how a number of self-associating surfactant systems including microemulsions could be used in drug delivery applications, and we will go on to discuss specific cases in the following sections of this review. All of the systems shown in Table 1 have the potential to protect labile compounds, but it is still the case that there are few examples of microemulsion-based drug delivery systems used in commercial drug formulations.

The attraction of o/w microemulsion systems lies in their ability to incorporate hydrophobic drugs into the apolar oil phase thereby enhancing their solubility [43–48,88,89]. However it is worth noting that most drugs are not especially soluble in hydrocarbon oils, rather the polarity of the majority of poorly water-soluble drugs favour their solubilisation in

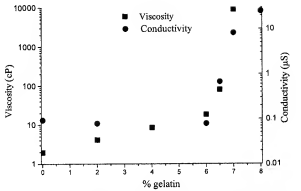

Fig. 9. Influence of the addition of gelatin to a water-in-dodecane microemulsion stabilised by AOT on viscosity and conductivity. [Water]/[Surfactant] molar ratio of 45 and AOT molarity of 0.15. Redrawn from [36].

small/medium molecular volume oils such as tributyrin or Miglyol 812. In fact formulating a drug in a hydrocarbon oil-in-water microemulsion may offer no advantage in terms of solubilisation over the corresponding micelle [92]. The dispersal of the drug as a solution in nanometre-sized droplets enhances the rate of dissolution into a contacting aqueous phase, and in vivo generally results in an increase in drug bioavailability. It is also noteworthy that the use of o/w microemulsions in drug delivery is more straightforward than is the case with w/o microemulsions. This is because the droplet structure of o/w microemulsions is often retained on dilution by a biological aqueous phase, thereby permitting oral as well as parenteral administration. However, the process of dilution will result in the gradual desorption of surfactant located at the droplet interface. This process is thermodynamically driven by the requirement of surfactant to maintain an aqueous phase concentration equivalent to its critical micelle concentration (CMC) under the prevailing conditions of temperature, pH and ionic strength. Because non-ionic surfactants typically have lower CMCs than their ionic counterparts, o/w microemulsion dosage forms based on non-ionic surfactants and designed for oral or parenteral use are likely to offer superior in vivo stability.

In contrast, the use of w/o microemulsions for oral or parenteral drug delivery is complicated by the fact that they are destabilised to a much greater

Table 1
Equilibrium phase structures encountered in oil–water–surfactant systems. From [46][a]

Micelles/swollen micelles/microemulsions
Reverse micelles/reverse swollen micelles/reverse microemulsions
Worm (polymer)-like micelles
Cubic/reverse cubic phases
Hexagonal/reverse hexagonal
Oil and/or water swollen lamellar phases

[a] In addition to the above, one phase structures, a range of two and three phase systems are observed, in which one of the above exists in equilibrium with an excess of oil and/or an excess of water.

extent when diluted by an aqueous phase. This is due to the increase in the volume fraction of the aqueous phase which increases the ratio of water to surfactant (ω_o) leading to droplet growth and eventually percolation. If the dilution continues, phase separation or inversion may occur and this will result in load dumping. However, there are advantages to be gained from formulating drugs in w/o microemulsion systems. Peptide drugs, for example, generally have little or no activity when delivered orally and are highly susceptible to proteolysis in the gastrointestinal tract [93]. Parenteral drug administration, especially for chronic conditions is not well accepted by patients and can lead to issues with compliance. Consequently, the oral delivery of labile drugs is the focus of growing attention, particularly as many of the new therapeutic agents in development are hydrophilic drugs such as peptides or oligonucleotides. Hydrophilic drugs of this kind can be successfully incorporated into the dispersed aqueous phase of w/o microemulsion droplets where they are afforded some protection from enzymatic degradation when administered orally [88]. In addition, the presence of surfactant and in some cases cosurfactant, for example medium chain diglycerides in many cases serves to increase membrane permeability thereby increasing drug uptake [88,89,94–98]. Drug delivery forms based on w/o microemulsions can also be employed where dilution by an aqueous phase is less likely to occur, such as after intramuscular injection [99]. Similarly, microemulsions and microemulsion gels have found application as topical agents where the surfactants and in some cases the oil phase itself act as penetration enhancers to facilitate transdermal drug delivery [85,90,91, 100,101].

Clearly then, in order to aid successful formulation, it is essential to study the phase behaviour of potential combinations of water, surfactant and oil. However, the studies can only ever act as a broad predictor of the likely fate of a microemulsion dosage form after delivery. For oral dosage forms in particular, it is not possible to realistically model the in vivo interactions of microemulsion components with the complex variety of food materials and digestive fluids present in the gastrointestinal tract.

4.1. Pharmaceutically acceptable excipients

Perhaps the most significant problem associated with formulating microemulsions is the difficulty associated with excipient acceptability. The vast majority of phase behaviour studies have been carried out using surfactants and oils, which do not have regulatory approval for use in pharmaceutical products. The majority of the work reported in the scientific literature concerns microemulsion systems based on either hydrocarbon oils such as heptane or dodecane, or cyclic oils such as cyclohexane. The most commonly used surfactants contain hydrophobes of 12 carbons such as sodium dodecyl sulphate (SDS) and tetraethylene glycol monododecyl ether ($C_{12}E_4$). Based on the results of these studies a number of workers have developed guidelines to aid in the formulation of microemulsions. However care must be taken when extrapolating 'guidelines' developed for such systems to pharmaceutically acceptable systems. For example it is widely reported that it is not possible to solubilise an oil which is larger than the hydrophobic chain length of the surfactant, yet recent studies have shown that using $C_{18:1}E_{10}$, it is possible to solubilise long chain triglyceride to a greater extent than its medium or short chain counterparts [42,92].

Nevertheless, quite a number of studies have deliberately chosen to employ naturally occurring excipients such as lecithins and glycerides, and there is even advice in the literature on the formulation of supposedly non-toxic microemulsions [102]. The regulatory status of the different excipients will also depend on their intended use. Thus while a reasonable range of surfactants would be deemed acceptable for use in topical formulations, the number considered safe for oral and especially parenteral use would be very restricted in comparison. This observation may go part way to explaining why a large number of researchers investigating microemulsions for drug delivery have concentrated on developing topically applied delivery systems. Many of the surfactants and oils that are regarded as acceptable are food grade materials or have a history of use in the pharmaceutical arena [103], for example as parenteral emulsion dosage forms [104]. There are a number of comprehensive studies in the literature concerning the preparation and characterisation of lecithin-based microemulsions [15,19–21,25,26, 52,56,59,65–69,87,89,97,99,105–125].

Non-ionic surfactants can be useful alternatives to naturally occurring surfactants, and polyoxyethylene sorbitan *n*-acyl esters (Tweens), for example, have been reported to have minimal toxicity. Although there are some restrictions, the use of polyoxyethylene sorbitan monooleate (Tween 80) and polyoxyethylene sorbitan monolaurate (Tween 20) appear acceptable for oral or parenteral use [126]. Importantly in some cases non-ionic surfactants such as the polyoxyethylene *n*-alkyl ethers (C_nE_m) are able to form microemulsions without the need for cosurfactant [127,128]. This is helpful as it reduces the complexity of the phase behaviour, and eliminates the requirement for inclusion of medium chain alcohols, since these cosurfactants have a poor toxicity profile. Furthermore, the insensitivity of non-ionic microemulsions to pH and electrolyte concentration relative to their ionic counterparts represents an added benefit. There are consequently a large number of formulation studies involving non-ionic surfactants as microemulsion excipients [18,38,86,92,127–133]. However, the biodegradability of many non-ionic surfactants raises issues with regard to long-term toxicity, especially in chronic use.

It is evident from the literature that there is a move toward the use of alternative 'safe' surfactants other than lecithin and the non-ionic surfactants named above. Thus there are a number of studies that have been conducted using *n*-alkyl amine *N*-oxides either in isolation or in combination with lecithin [42,114]. Interestingly these surfactants are much more rapidly biodegradable than the polyoxyethylene *n*-alkyl ethers. Furthermore there has also been a recent report detailing the potential of a biodegradable version of the amine *N*-oxide surfactant, dimethyldodecylamine *N*-oxide, for use in microemulsion formation [134]. Sugar surfactants such as alkyl glucosides have also received a good deal of attention [64,135–140]. Interestingly while sugar surfactants are widely considered as biodegradable they still exhibit a level of haemolytic activity on a par with that exhibited by the polyoxyethylene *n*-alkyl ethers. The use of sucrose fatty acid esters as surfactants in the stabilisation of microemulsion phases has also been investigated [75,85,141–144]. Surfactants based on polyglycerol fatty acid esters have been suggested as 'orally safe' in a study designed to investigate the potential of microemulsions for the delivery of protein drugs [145]. Monoglyceride has also been used as surfactant in its own right to stabilise a triglyceride-in-water microemulsion system [83], and by Constantinides and coworkers [96,146] in a self-microemulsifying drug delivery system (SMEDDS).

In many cases, a requirement for cosurfactant causes difficulty in the formulation of acceptable microemulsions because the majority of studies have chosen to employ medium chain length alcohols as the cosurfactant of choice. Unfortunately, there are significant toxicity and irritancy issues with these materials, which preclude their use in pharmaceutical formulations. In addition, the aqueous solubility of cosurfactants in o/w microemulsion systems is often higher than that of the principal surfactant. Consequently, when the o/w microemulsion is diluted, the cosurfactant partitions more strongly to the aqueous phase. This has the effect of depleting the cosurfactant concentration at the oil/water interface, thereby destabilising the microemulsion droplet. Alternatives to medium chain alcohols have been evaluated such as short chain amines [147] and alkanoic acids [21], however these cosurfactants behave in much the

same way as the alcohols and toxicity remains an issue. A number of small, relatively polar molecules are also thought to act as cosurfactants. Ethanol, for example, has been used as a 'cosurfactant' in both o/w and w/o microemulsion systems. Polyhydric alcohols such as sorbitol and sucrose have also been used as additives to facilitate microemulsification but the resulting o/w formulations were unstable on dilution with water as the aqueous solubility of these materials is high [86,148].

The utility of ionic surfactants is also relatively limited in pharmaceutical dosage forms, however the use of ionic surfactants in formulation studies has been reported on a number of occasions [18,33]. Coformulation of the anionic surfactant AOT with a number of different drugs as a microemulsion dosage form has been more widely reported. Examples, which have appeared in the literature, include vitamin K and steroids such as hydrocortisone, prednisolone and betamethasone [34,106]. The solubilisation of pilocarpine and chloramphenicol was looked at in both AOT and DDAB w/o microemulsions [31], whilst the trandermal delivery of glyceryl trinitrate has been tested in vivo from a variety of related AOT systems [149]. As has been indicated previously, the advantage of twin tailed surfactants like AOT and DDAB is that w/o microemulsions form easily without the aid of cosurfactant. However, when AOT microemulsions are formulated in eombination with non-ionic surfactants, the extent of the microemulsion region is often markedly increased, and in addition the temperature range over which the microemulsion is stable is generally improved [18,33,43]. Fig. 10 gives an example of the increase in microemulsion area of existence achievable in a mixed AOT-non-ionic surfactant system over that seen when either surfactant is used alone.

Total parenteral nutrition emulsions have provided a starting point from which suitable oils might be selected for formulation into pharmaceutically acceptable microemulsions. Medium chain triglycerides such as Miglyol 812 have been used quite frequently [42,128,148], but fatty acid esters are also a popular choice. Of these, isopropyl myristate (IPM) is the most popular [19,21,34,38,66,67, 69,71,74,86, 90,106,115,117–120,123,124,141,150–154], but isopropyl palmitate (IPP) has also found application [150,155], especially in microemulsion gels [100,151,156,157]. Ethyl or methyl esters of lauric, myristic and oleic acid have also been employed as pharmaceutical excipients for microemulsion formulation [63,92,99,108,151]. Ryan and Kaler have employed alkyl ethylene glycol ethers such as ethylene glycol diethyl ether and ethylene glycol dibutyl ether as the oil phase. The combination of these rather hydrophilic oil phases with alkyl glucoside surfactants enhances the surfactant solubility in the oil phase and microemulsification can be achieved without the need for cosurfactant [137,139]. However, with one or two exceptions it is clear that the majority of oils intended for pharmaceutical use are large and semi-polar, and therefore rather different to the alkane oils most commonly reported in the scientific literature.

4.2. Low viscosity microemulsion systems

At relatively low dispersed phase volume fractions, the microemulsion generally contains nanometre-sized droplets of oil or water. If the droplets are non-interacting the resulting microemulsion will be low viscosity and may therefore be suitable for oral, parenteral, pulmonary or even ocular delivery. The potential of these systems for use topically has not been overlooked, however it is recognised that more viscous microemulsion systems are preferred for this application. Suitable systems would include those containing extended structures such as cylindrical or polymeric worm-like or micelles [52,59] or which have been thickened through the addition of a specific gelling agent [90,91]. Cubic or hexagonal mesophases identified in the bicontinuous microemulsion region may also be of sufficiently high viscosity to have application as topical delivery systems [38,85,142].

Oil-in-water microemulsions have been used to solubilise steroidal drugs such as prednisolone, hydrocortisone and betamethasone, testosterone and its esters and progesterone [34,92,106,127]. Interestingly, it has been noted that hydrophobic drugs need to have a significant solubility in the dispersed oil phase for the o/w microemulsion system to offer a marked benefit over the micellar system alone [127]. However, the oils in which the drug is most soluble do not necessarily form microemulsions with the highest drug solubilisation capacity [92].

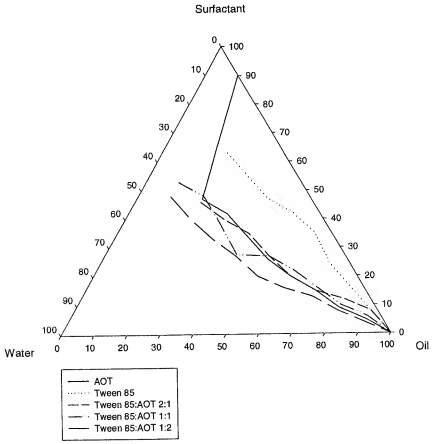

Fig. 10. Partial ternary phase diagram for Tween 85 (dot) and AOT (solid) alone, or as a surfactant mixture in combination with isopropylmyristate (oil) and H_2O at 298 K. Compositions to the right of the phase boundary are optically clear, single phase, water-in-oil microemulsions. The weight ratios of Tween 85:AOT employed were 2:1 (dash), 1:1 (dash-dot-dot) and 1:2 (long dash). [S. Kantaria, G.D. Rees, M.J. Lawrence, unpublished observations.]

The utility of w/o microemulsions for the sequestration of labile peptide drugs was demonstrated using an analogue of luteinizing hormone-releasing hormone (LHRH) to control testosterone levels in rats [99]. The LHRH was solubilised in a lecithin-stabilised system with ethyl oleate as the oil phase and hexanoic acid as cosurfactant, and was administered as an intramuscular injection.

The peroral delivery of insulin, cyclosporine A and vasopressin from microemulsion formulations has been reviewed by Ritschel [158]. Interestingly, some drugs such as diclofenac diethylamine are amphiphiles in their own right and indeed this drug self assembles under aqueous conditions to yield a liquid crystal phases. Coformulation of diclofenac diethylamine with phospholipid, not unexpectedly, gives rise to some complex phase behaviour, and microemulsion phases have been identified from the ternary phase diagram [159]. Diclofenac release and permeation through excised human skin was subsequently reported [112] with the researchers clearly demonstrating the importance of the nature of the phase structure of the amount of drug released (Fig. 11). Cholesteryl ester prodrugs of ibuprofen and flufenamic acid have also been incorporated into phospholipid microemulsions. In this case the dispersed oil phase was modified by the addition of cholesteryl oleate [109].

An interesting non-aqueous microemulsion formulation based on lecithin/glycerol/soybean oil has

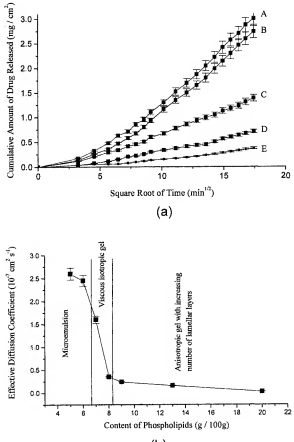

Fig. 11. (a) Release of diclofenac diethylamine from vehicle with 5% diclofenac diethylamine and various concentrations of phospholipids at 20°C. (A,B) Microemulsions with (A) 5% phospholipids and (B) 6% phospholipids; (C–E) anisotropic gel with (C) 9.3%, (D) 13.8% and (E) 20% phospholipids, respectively. (b) Effective diffusion coefficient of diclofenac diethylamine from vehicle with 5% diclofenac diethylamine vs content of phospholipids. Data represent mean and standard deviation of three duplicate determinations. Redrawn from [112].

been reported. The formulation was designed for parenteral delivery and contained the octyl ester of ibuprofen. Modification of the microemulsion formulation with a poloxamer alters the absorption process with the result that the drug is targeted to reticuloendothelial system-rich organs [113].

A novel pressurised aerosol system has been devised for the pulmonary delivery of salbutamol using lecithin-stabilised microemulsions formulated in trichlorotrifluoroethane [15]. The ocular use of microemulsion systems has also been indicated and the β-blocker levobunolol has been incorporated into o/w microemulsions with this purpose in mind [107]. Pilocarpine has also been incorporated into a microemulsion designed for ocular use, which was stabilised by sucrose fatty acid ester surfactants [141]. Poloxamer-stabilised microemulsions containing triacetin, castor oil, water and propylene glycol have also been formulated as potential ophthalmics. Formulations were prepared over a range of compositions, and in vitro drug permeation studies were successfully conducted using indomethacin, diclofenac and chloramphenicol [160].

Antifungals including clotrimazole, cyclopiroxolamine and econazole nitrate have been incorporated into IPM and IPP microemulsions designed for topical use and stabilised by Tween 80 [150]. Azelaic acid, a drug used in the treatment of pigmentary disorders, has also been investigated as a topical microemulsion formulation. In this case the azelaic acid was solubilised in a viscosised o/w microemulsion and tested in a hairless mouse model where flux enhancements were observed [161]. The local anaesthetic pentacaine has been incorporated into a w/o microemulsion and subjected to in vivo testing on rabbits [162].

Interestingly, microemulsions have not only been used directly as drug delivery vehicles, but have also been used indirectly as a means of producing solid drug-loaded nanoparticles. In this case, solid lipid nanoparticles were synthesised by spraying an o/w microemulsion into cold water at a temperature below the melting point of the oil. The microemulsion itself is typically stabilised by lecithin and uses a mixture of capric and palmitic acid as the oil phase. The microemulsion in addition may contain taurodeoxycholate and alkyl phosphates as cosurfactants. The resulting drug loaded particles could be isolated, washed and freeze dried [163–169]. The properties of w/o microemulsions were similarly exploited by utilising droplets as supermolecular templates for the synthesis of drug-loaded polybutylcyanoacrylate nanoparticles [170,171].

4.3. High viscosity systems and microemulsion gels

A bicontinuous microemulsion stabilised by a mixture of non-ionic surfactants was designed for the

topical delivery of the local anaesthetic lidocaine [38]. Comparative studies have shown that an o/w microemulsion gel containing the analgesic anti-inflammatory flufenamic acid and stabilised by PEG-7 glyceryl coccoate (Cetiol HE) outperforms the corresponding macroemulsion, hydrogel and cream. The formulation was tested in vitro and in vivo by topical administration on rats [172].

The interaction of cylindrical or worm-like micelles in microemulsion formulations tend to gives rise to high viscosity systems. The proposed structure of a worm-like (or spaghetti-like) micelle is shown in Fig. 12. Lecithin in particular is known to form such systems in microemulsions at low water content and the utility of the resulting organogels as novel matrices for the transdermal transport of drugs has been investigated [100]. Scopolamine, broxaterol and propranolol have been incorporated into lecithin organogels based on either cyclohexane, isooctane or IPM. Ten-fold enhancements of permeation rates through excised human skin were reported compared to solvent only controls [100,173]. Aromatic tetraamidines with antitumour activity have also been incorporated into lecithin organogels for transdermal drug delivery [151]. The in vivo efficacy was determined in nude mice carrying xenografted tumour cells and was considered to be encouraging. Methyl nicotinate has been formulated in a lecithin IPM organogel and tested in vivo with human subjects [174]. The utility of these lecithin organogels has been supported by a human skin irritation study, which showed a very low irritancy potential for the soybean lecithin/IPP/water system [156,175].

4.4. SMEDDs

Self-emulsifying drug delivery systems (SEDDS) and SMEDDS can be described as isotropic solutions of oil and surfactant, which form o/w (micro)emul-

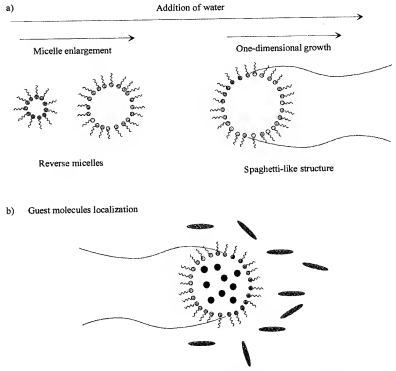

Fig. 12. Microemulsion-based lecithin gels. (a) schematic representation of the formation of lecithin gels upon addition of water to small lecithin reverse micelles in apolar solvents. (b) localisation of solubilised 'guest' molecules within the lecithin gels. Lipophilic drug (stripped bar); hydrophilic drug (black circle); and amphiphilic drug (shaded head with attached tail). Redrawn from [209].

sions on mild agitation in the presence of water [176,177]. The utility of SEDDS has been investigated by Charman and coworkers who, although unable to show enhanced bioavailability of an investigational lipophilic drug WIN 54954, were able to demonstrate greatly improved pharmacodynamics using systems based on medium chain triglyceride (MCT) and ethoxylated glyceryl trioleate (Tagat TO) [178]. More recently, self-emulsifying w/o microemulsions based on MCTs such as Captex 355 and Captex 8000 have been reported. The systems contained a mixture of mono and diglycerides (Capmul MCM) in combination with Tween 80 as surfactant. The bioavailabilities of calcein, a water-soluble marker, and an RGD peptide were shown to be significantly increased using a microemulsion concentrate and preformulated w/o microemulsions compared to the control aqueous formulation [96,97,146]. The bioavailability of a poorly water soluble 5α-reductase inhibitor has similarly been shown to be improved in Beagle dogs [179]. It is also notable that the presence of liquid crystalline phases in the pseudo binary oil/surfactant mixtures are claimed to be a feature of the most efficient SEDDS [180].

The most notable example of a SMEDDS relates to the oral delivery of cyclosporin A, in particular the commercial Neoral® formulation. Cyclosporin A is a cyclic undecapeptide used as an immunosuppressant in transplantation surgery, and in contrast to most peptide drugs is hydrophobic. The original Sandimmune® formulation was based on a solution of cyclosporin A in vegetable oil. Although the coadministration of triglyceride with the cyclosporine A improved its bioavailability, there was considerable pharmacodynamic inter- and intra-patient

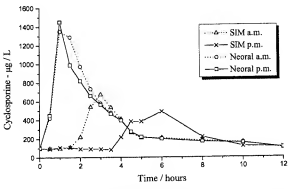

Fig. 13. Representative cylcosporine blood concentration profiles from a renal transplant patient given the currently marketed formulation Sandimmune® (SIM) or the new formulation (Neoral) without food (a.m.) or with food (p.m.). Redrawn from [184].

variability as can be seen in Table 2 and Fig. 13. This variation can be ascribed to the proposed mechanism of uptake in vivo which is considered to be related to the lipolysis of the triglyceride yielding lower partial glycerides which then act as emulsifiers and enhance drug uptake [181]. The Neoral® formulation uses an isotropic concentrated blend of surfactant based on medium chain length partial glycerides, a medium chain length triglyceride oil and drug. Exposure of this concentrate to water results in formation of initially a w/o microemulsion which on further mixing with water undergoes phase inversion to yield an o/w microemulsion. The delivery of cyclosporine A via microemulsion formulations has been considered in some detail [158,182,183], and the superiority of the Neoral® microemulsion preconcentrate over the original Sandimmune® formulation has been demonstrated on several occasions [184–186]. Furthermore, the available data has been

Table 2
Mean (CV%) pharmacokinetic parameters following twice-daily dosing with Sandimmune® (SIM) or Neoral® by 11 stable renal transplant patients. From [184][a]

Parameter	SIM fasting	SIM non-fasting	Neoral fasting	Neoral non-fasting
T_{max} (h)	2.1 (33.3)	2.6 (76.9)	1.5 (33.3)	1.2 (33.3)
C_{max} (µg/l)	663 (34.5)	528 (40.5)	997 (20.0)	892 (35.8)
C_{min} (µg/l)	78 (30.8)	92 (29.3)	94 (22.3)	100 (23.0)
AUC (µg-h/l)	2645 (25.7)	2432 (24.3)	3454 (17.6)	3028 (19.7)
PTF%	261 (23.4)	212 (36.8)	317 (18.0)	309 (31.1)

[a] All concentrations measured in whole blood at steady state.
AUC was measured over a dosing interval. *PTF*% = percentage peak–trough fluctuation.

reviewed by Noble and Markham who confirm the conclusion that Neoral® offers more predictable and more extensive drug absorption than the standard Sandimmune® formulation [187].

Interestingly, it has been noted that the bioavailability of vasopressin and insulin from w/o microemulsions is higher when the oil phase is based on straight-chain rather than branched-chain fatty acid esters [182]. It is likely that the observed differences in bioavailability are related, at least in part, to the large reduction in lipolytic activity exhibited by lipases toward branched chain fatty acid substrates [188]. After administration, the microemulsion formulated with straight chain fatty acid esters will undergo rapid enzymatic hydrolysis being degraded in the gastrointestinal tract. The breakdown products are surface active and will stabilise any (micro)emulsion that may form, as well as acting as membrane permeation enhancers [189]. As a consequence of the important role played by metabolic processes in vivo, formulators should be aware that certain hydrophilic surfactants such as Brij 96/Brij 97 ($C_{18:1}E_{10}$), Tween 80 and polyoxyethylene 40 hydrogenated castor oil (Cremophor RH40) have been shown to inhibit lipolysis in vitro. Clearly if this behaviour is mirrored in vivo one of the principal mechanisms facilitating drug uptake would be compromised. It is therefore encouraging to observe that co-incubation of these hydrophilic surfactants with lipophilic surfactants such as oleic acid or medium chain length partial glycerides reverses the inhibition [181].

It is also notable that in the case of w/o microemulsion systems, there is no obvious correlation between droplet size and oral bioavailability. This contrasts with the known relationship between o/w emulsion droplet size and bioavailability [190,191]. Ignoring the problems of sizing such concentrated systems, this behaviour is expected since the w/o microemulsion phase inverts or phase separates after administration, thereby releasing any drug previously compartmentalised in the aqueous droplet core. Where differences in bioavailability would be predicted for w/o systems are occasions where surfactant selection is altered. Alternatively, and as has just been discussed, changes in bioavailability may occur where there are significant differences in the biodegradability of formulation components which might lead to the production or otherwise of additional surfactant species.

An interesting SMEDD variant has been employed for the dermal delivery of β-blockers in an anhydrous surfactant/IPM vehicle [155]. After topical administration with an occlusive patch, water partitions into the SMEDD forming a microemulsion gel phase. As the extent of hydration increases, the drug solubility decreases resulting in a supersaturated system which the authors claim results in enhanced drug activity and improved pharmacodynamics [192]. In a related report, organogels were prepared from lecithin and a fatty acid ester oil containing indomethacin [193]. This pseudo binary system was effectively anhydrous but in common with the previously mentioned study, water uptake would be expected to result in the formation of the standard lecithin microemulsion gel which has been previously discussed. As expected, enhanced permeation rates were reported for the indomethacin gel in a hairless mouse model.

5. Recent developments and future directions

For the purposes of this review, recent developments will for the most part constitute an evaluation of the literature in the area of microemulsions and microemulsion-based systems for drug delivery for the period beginning 1997 to date.

5.1. Component selection

It is notable that an increasing proportion of new studies recognise the benefit associated with employing as far as possible, pharmaceutically acceptable surfactants, cosurfactants and oils [194]. Naturally occurring surfactants and oils remain an attractive option, and the phase behaviour and microstructure of microemulsions based on soybean phosphatidylcholine and triglycerides has recently been reported, although propanol was used as the cosurfactant [65]. The addition of cosurfactants such as alkanol phosphocholines have been shown to increase the extent of the microemulsion region in lecithin/triglyceride systems [120]. However the regulatory status of these particular alkanol phosphocholines, which were synthesised 'in house' is unclear, and once again

short chain alcohols including ethanol, propanol and butanol were used as cosolvents/cosurfactants. The same group has also reported on the phase behaviour of systems containing lecithin and 2-acyl lysolecithin derivatives coformulated with medium chain triglycerides or IPM as the oil phase and ethanol as cosurfactant/cosolvent. These modified phospholipids also increase the extent of the microemulsion region most probably by increasing the fluidity of the surfactant film [124]. Significantly, most of the work investigating the formation of lecithin-based microemulsions has reported an improvement in the extent of the w/o region. In contrast, relatively little success has been achieved in the water-rich part of the phase diagram. However a recent study describes the formation of o/w lecithin-based microemulsions in combination with amine *N*-oxide surfactants [195].

5.2. Phase behaviour

The phase behaviour of systems stabilised by nonionics other than polyethylene glycol ethers continues to be a prominent area of research activity. A number of phase behaviour studies involving dermatologically acceptable sugar surfactants such as alkyl glucosides have appeared. As has been indicated previously, the low solubility of sugar esters in hydrophobic alkane oils generally necessitates the use of a cosurfactant such as a medium chain alcohol in order to facilitate microemulsification. However, unlike their polyoxyethylene surfactant counterparts, surfactants based on sugar esters are much less sensitive to temperature [74]. Until recently it was not possible to prepare microemulsions stabilised by alkyl glucosides in the absence of a cosurfactant. However it has recently been reported, that by employing a polar oil phase based on alkyl ethylene glycol ethers, in which the alkyl glucosides are more soluble, it is possible to prepare glycoside stabilised microemulsions in the absence of cosurfactant [137–139]. An interesting alternative approach in microemulsions stabilised by octyl monoglucoside has been to employ geraniol, a non-toxic perfume alcohol ($C_{10}H_{17}OH$) as a cosurfactant/cosolvent [140]. As is the case with addition of alcohols or alkyl glycerol ethers to alkyl glucosides, the addition of geraniol allows phase inversion of the system to occur. Microemulsions formulations stabilised by sucrose monolaurate and sucrose dilaurate containing ethyl and cetyl 2-(hexylethyl)-2-hexanoate as the oil phase and diethyleneglycol monoethyl ether as cosurfactant have also been reported. These systems were characterised using FFEM, SANS and viscosity [75]. The formation of microemulsion and liquid crystal phases in biocompatible sucrose alkanoate systems has also been claimed [61]. Very recently, a study has appeared examining the water solubilisation capacity of microemulsions stabilised by a variety of sucrose esters including mono, di and polyesters of lauric, palmitic, stearic and oleic acid. Medium chain triglyceride was employed as the oil phase, however the study employed medium chain alkanols as cosurfactants [144].

The influence of drug incorporation on the phase behaviour and structure of microemulsion system based on water, propanol, lecithin and medium chain triglyceride has also been investigated [125]. The preparation of non-toxic microemulsions has been described for mixtures of IPM or orange oil with lecithin. Notably, alkane diols such as pentane-1,2-diol and octane-1,2-diol were employed as possible non-toxic replacements for propanol and pentanol, respectively [119]. Interestingly in their study involving the use of neural networks to predict microemulsion phase behaviour Richardson and coworkers predicted that 1,2 hexanediol would be a suitable surfactant for the production of balanced lecithin-based microemulsions [196]. In terms of drug solubilisation capacities, microemulsions should fare better than micelles because of the extra locus for solubilisation provided by the oil phase. However, a recent phase behaviour study has shown that whilst polar oils offered better drug solubility, the solubility of the drug in the microemulsion itself was also determined by the extent to which low molecular volume oils penetrate the PEO region and destroy drug solubilisation sites [92].

5.3. Block copolymer micelles and microemulsions

It is noticeable from the literature that there has been a significant increase in the number of papers describing the use of block copolymer systems for drug delivery [197–204]. Given that both block copolymers and conventional surfactants are am-

phiphilic, it is unsurprising that there is great similarity between the self-association and adsorption properties of these two classes of compounds. However, an advantage of amphiphilic block copolymers over conventional surfactants is the relative ease with which the physicochemical properties can be 'tailored' to suit a given application. Specific examples might include the glass transition temperature (Tg) of the hydrophobic core and the CMC [197,200]. In addition, amphiphilic block copolymers may be less haemolytic than conventional surfactants [201]. As is the case with conventional surfactants, the addition of oil or water respectively to block copolymer micelles or reverse micelles can lead to the formation of o/w or w/o microemulsion systems, respectively. The physicochemical characterisation of block copolymer microemulsions has been reported, and some of their potential applications examined [205,206]. However, their formulation and potential application in drug delivery has received little attention to date with the exception of a study by Siebenbrodt and Keipert which examined the formulation of Poloxamer-stabilised microemulsions containing triacetin and castor oil, water and propylene glycol over a range of compositions [160].

Block copolymer microemulsions are likely to become the focus of more intense research over the next few years, particularly where drug delivery applications are concerned. Mechanistically, the dynamics associated with such systems have much in common with conventional microemulsion stabilised by non-polymeric surfactants. However, the timescales associated with molecular events such as unimer insertion or exchange can be several orders of magnitude slower in the case of block copolymers [207]. The aforementioned differences would be expected to have considerable impact on microemulsion stability and the control of drug release in vivo.

5.4. Comparative drug delivery studies

A number of comparative reports have appeared in the literature, some of which have evaluated the utility of microemulsion formulations against alternative delivery systems including liposomes, micelles, emulsions and creams [208,209]. Regrettably, the picture is often complicated as the same components are not always employed in the production of the various systems under test. Nevertheless, the antitumour drug camptothecin, for example, has been tested in vitro for anti-proliferative activity toward cultured human leukaemic K 562 cells as a micellar solution, a microemulsions and incorporated into liposomes. Activities in all cases were moderately enhanced over the control. Isostearyl isostearate was employed as the oil phase and the microemulsion stabilised by a mixture of Labrosol® and Plurol isostearate® [208]. The peroral delivery of parasympatholytic trospium chloride has been compared using two w/o microemulsion formulations as well as a cyclodextrin formulation and an aqueous solution control. The trospium carries a quaternary nitrogen and is positively charged which may play a role in its poor peroral bioavailability (3 to 11%). Uptake of trospium may also be inhibited by formation of insoluble complexes with acidic residues of mucopolysaccharides. This would be consistent with the observation that ion pair formation increases its lipophilicity and improves bioavailability. Disappointingly, in this particular study, bioavailabilities were either equivalent or lower than aqueous solution control [210]. Bicontinuous microemulsions based on sucrose monolaurate or sucrose dilaurate have been used for the topical delivery of niflumic acid, a potent anti-inflammatory. The 1% microemulsion was as effective as the 3% commercially available niflumic aeid ointment [85]. An in vitro comparative study of the percutaneous absorption of propranolol from an emulsion, an o/w microemulsion and a micellar solution stabilised by Tween 80 and Span 80 has also been reported. Although the o/w microemulsion was superior to the emulsion system, both of which contained IPM, the micellar delivery system containing propranolol was the most effective [152].

5.5. Oral delivery — SEDDs and SMEDDs

The formulation of w/o microemulsions for use as SEDDS or SMEDDS has been investigated using blends of low and high HLB surfactants, which were commercially available and pharmaceutically acceptable, typically sorbitan esters and Tween 80. The oil phase comprised long or medium chain length glycerides [73]. The microemulsions were characterised by a variety of techniques including conduc-

tivity, viscosity and photon correlation spectroscopy (PCS). In a related study, an optimised o/w microemulsion formulation for the delivery of cyclosporin A prepared using Cremophor EL® as surfactant, Transcutol® as a cosurfactant and Captex 355® as the oil phase has been reported. Bioavailability enhancements of 3.3 and 1.25 were observed relative to the Sandimmune® and Sandimmune Neoral® formulations [211].

The formulation of SEDDS has also been recently reviewed and a number of pertinent observations made [98]. Concerns have been raised for example regarding administration of dosage forms containing high concentrations of surfactant, for example, and opportunities to evaluate chronic toxicology are limited in comparison to tablet formulations [95]. As has been indicated already, the presence of water or another polar cosolvent in a SEDDS formulation may mean that the concentrate is itself a microemulsion. In highlighting factors that predispose efficient microemulsions the authors note that self-emulsification requires least energy close to the PIT, which is also where the capacity for water solubilisation is enhanced. Mechanisms of self-emulsification discussed include the dynamic formation of liquid crystalline units at the oil/water interface, at least for simple SEDDS for example based on Tween 85, a medium chain triglyceride and drug [98]. For more complex SEDDS, a mechanism of 'diffusion and stranding' producing fine emulsions or microemulsions may also operate. The formulation of a SMEDDS containing flurbiprofen and based on phospholipid, ethyl oleate and ethanol has also been reported. After parenteral administration, significant increases in flurbiprofen half-life and an altered biodistribution pattern were observed [122] as is clearly illustrated in Fig. 14. In another highly pertinent study, the influence of phase transformation on indomethacin release from microemulsions is examined [123]. The systems of interest were prepared from water (42%), IPM, lecithin, lysolecithin and ethanol. The compositions were selected such that emulsions, microemulsions, or liquid crystal were formed on dilution. With the exception of phase transformations to liquid crystals, the release rate of indomethacin was considered by the authors to be too rapid for controlled drug delivery. However, the release rate of drugs in general from these systems

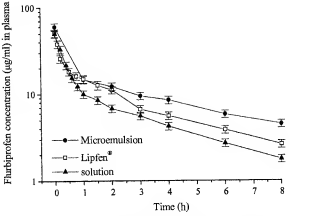

Fig. 14. The plasma concentration–time profiles of flurbiprofen after intravenous administration of flurbiprofen solution, commercial product (Lipfen®, flurbiprofen axetil-loaded emulsion) and flurbiprofen-loaded microemulsion (EO: lecithin: DSPE–PEG: flurbiprofen = 8:3:1:2) equivalent to 2.5 mg/kg as flurbiprofen to rats ($n = 5$). Redrawn from [122].

will also depend on the oil/water partition coefficient and factors such as droplet size and specific drug-excipient interactions. Compositional variables have been shown to affect the in vitro release of drug from microemulsion formulations in a number of other cases [53,71].

A pharmacokinetic study with the Sandimmune cyclosporin A Neoral® microemulsion concentrate exhibited the expected improvements in bioavailabilty and inter/intra-patient variability and was shown to facilitate the effective management of psoriaris [212]. Very recently, pharmacokinetic trials on microemulsion formulations for the oral administration of Neoral and a macrolide immunosuppressant SDZ RAD have been evaluated in non-human primates and were shown to have promising efficacy and tolerability [213]. Valsopdar, a P-glycoprotein modulator and an analogue of cyclosporine, has also been tested as an oral SEDDS formulation both free and in gelatin soft-capsules in a clinical trial against an i.v. infusion of the drug. Absolute bioavailability was 60% and the rate and extent of drug absorption comparable to that of the IV infusion [214,215]. The formulation and performance of SEDDS containing the anti-malarial halofantrine has also been reported. The SEDDS and SMEDDS were isotropic mixtures of medium or long chain triglyceride, monoglyceride

(Capmul MCM), drug and ethanol. Six- to eight-fold improvements in bioavailability were observed relative to tablet formulations [216].

5.6. Parenteral, pulmonary and ocular delivery

The preparation and evaluation of flurbiprofen-loaded o/w microemulsions is one of the few recent reports of delivery systems designed for parenteral use. The systems of interest were prepared using ethyl oleate as the oil phase and Tween 20 as surfactant. The drug solubility was eight times higher than in buffer, but there was no significant difference in the pharmacokinetics after administration in rats [217]. Pharmaceutically acceptable microemulsions designed for i.v. administration have recently not only been formulated and characterised, but also tested in vivo for hemodynamic response [68]. The microemulsions comprised Miglyol 810N (MCT), soybean phosphatidylcholine (Epicuron 200), PEG 400, poly(ethylene glycol)(660)-12-hydroxystearate and ethanol. PFG–NMR indicated that the microemulsions formed over a range of compositions were bicontinuous, even at high oil concentrations. After administration, the bicontinuous microemulsions form o/w emulsions on dilution. In vitro studies showed the resulting droplets were small, with mean radii typically in the range 60–200 nm. Solubilisation studies using felodipine (a calcium antagonist) and an antioxidant H 290/58 were conducted, however in vivo studies were performed using the drug-free systems. The i.v. administration of the microemulsion formulations was performed by infusion into conscious rats over a 5-min period. Doses up to 0.5 ml/kg had no significant effect on acid–base balance, blood gases, plasma electrolytes, arterial blood pressure or heart rate [68].

The formulation of a water-in-HFA propellant microemulsion stabilised by fluorocarbon non-ionic surfactant and intended for pulmonary delivery has been described [16]. To date these workers have only presented the phase behaviour of the systems together with some light scattering results to prove the formation of a microemulsion. As yet no data has been reported on the incorporation of drug into these systems.

The development and characterisation of o/w microemulsions designed for ocular use has recently been reported [118]. The microemulsions containing pilocarpine were formulated using lecithin, propylene glycol and PEG 200 as cosurfactants, and IPM as the oil phase. The formulations were low viscosity fluids with a refractive index lending themselves to ophthamological application. The test microemulsions were non-irritant in rabbit eyes or hen egg membrane. A prolonged pharmacological effect was observed in vivo compared to the drug administered as a simple aqueous solution. This may have been related to increased bioavailability or enhanced retention or both. However, prolonged release was not observed in vitro using a cellulose membrane as permeability barrier.

5.7. Topical delivery

A number of recent reports detail microemulsion formulations designed for topical or transdermal application [218,219]. Both o/w and a w/o microemulsions have been evaluated in a hairless mouse model for the delivery of prostaglandin E1. The microemulsions were based on oleic acid or Gelucire 44/14 as the oil phase and were stabilised by a mixture of Labrasol (C_8 and C_{10} polyglycolysed glycerides) and Plurol Oleique CC 497 as surfactant [219]. Although enhanced delivery rates were observed in the case of the o/w microemulsion, the authors concluded that the penetration rates were inadequate for practical use from either system. The use of lecithin/IPP/water microemulsion for the transdermal transport of indomethacin and diclofenac has also been reported. Fourier transform infra red (FTIR) spectroscopy and differential scanning calorimetry (DSC) showed the IPP organogel had disrupted the lipid organisation in human stratum corneum after a 1 day incubation [157].

The transdermal delivery of the hydrophilic drug diphenhydramine hydrochloride from a w/o microemulsion into excised human has also been investigated. The formulation was based on combinations of Tween 80 and Span 20 with IPM. However two additional formulations were tested containing cholesterol and oleic acid, respectively. Cholesterol increased drug penetration whereas oleic acid had no measurable effect, but the authors clearly demonstrated that penetration characteristics can be modulated by compositional selection [153]. Com-

positional effects have also been investigated for the skin permeation of felodipine, a calcium antagonist, from o/w microemulsions stabilised by a surfactant mixture containing Tween 20 and taurodeoxycholate. IPM was used as the oil phase with benzyl alcohol as cosurfactant [90]. Characterisation of lecithin/alkanol/dodecane/water systems containing lidocaine and designed for topical use has shown the presence of L1 (micelles) and L2 (reverse micelles) isotropic droplet phases and a bicontinuous phase is the mesophase between L1 and L2 [121]. A combination of drug delivery strategies has been employed in a recent report describing the formulation of an inclusion complex of the anti-inflammatory piroxicam with β-cyclodextrin in an o/w microemulsion. Again designed for topical use, the microemulsion system contained IPM and was stabilised by the cationic surfactant hexadecyltrimethylammonium bromide [154]. In a related study this time employing an anionic surfactant, the transdermal permeation of glyceryl trinitrate through mouse skin was found to be enhanced by around a factor of around 10 by formulating the drug in AOT micelles and reverse micelles and w/o microemulsions. Irritation studies showed that the normal micelles caused moderate irritation but that AOT reverse micellar solutions caused little or no erythema [149].

An interesting application of gelatin microemulsion-based organogels (MBGs) which exploits the presence of surfactant-stabilised conducting aqueous channels has been their use in the iontophoretic transdermal delivery of a model hydrophilic drug. One of the structures proposed for these MBGs is given in Fig. 15. The MBGs were prepared using a variety of pharmaceutically acceptable surfactants and oils including Tween 80 and IPM [91]. Novel sorbitan monostearate organogels have also been prepared from vegetable oils and IPM. Prepared at elevated temperatures and then cooled, the surfactant self-assembles into inverse vesicles and then rod-shaped tubules. The organogels are opaque and thermoreversible, and may have been suggested as novel delivery vehicles for drugs and antigens [220]. The inclusion of a water component allows the formulation of w/o organogels which in common with gelatin MBGs, may have percolative electroconductive channels and can be used to solubilise hydrophilic drugs and vaccines as well as hydrophobic materials in the continuous oil phase [221]. Prolonged release of bovine serum albumin from a sorbitan monooleate w/o organogel injected intramuscularly to mice has been observed [100].

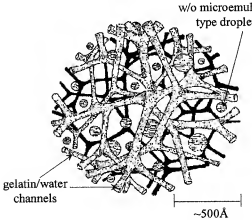

Fig. 15. Proposed microemulsion-based gel (MBG) structure based in small angle neutron scattering [237].

5.8. New developments

Environmentally responsive drug delivery systems are an interesting development and phase changes that occur after administration triggered by changes in temperature, pH or ionic strength can be particularly useful. One example of such behaviour involves the phase transformation of a reverse micellar solution of lecithin in IPM to a lamellar liquid crystal. In this case the transition was triggered by contact of the reverse micellar solution with a biological aqueous phase, resulting in the controlled release of the anti-inflammatory fenoprofen [222]. Very recently, similar behaviour has been exploited by the use of thermosetting microemulsions as delivery systems for periodontal anaesthesia. In this case, a block copolymer liquid microemulsion containing lidocaine and prilocaine was designed to form a gel after in vivo administration to the periodontal pocket [223]. Thermoresponsive polymeric block copolymer micelles based on poly(*N*-isopropylacrylamide) and poly(butylmethacrylate) and containing adriamycin have also been reported

[202]. The abovementioned fluid–gel transitions might be usefully exploited in an injectable fluid dosage form, which undergoes subcutaneous gelation after administration.

An interesting phase behaviour study has been conducted using mixtures of water, glyceryl monoleate and Pluronic F-127, the latter being a triblock PEP–PPO–PEO copolymer. A phase resembling a cubic bicontinuous microemulsion was identified which could be dispersed in water, and after microfluidisation yielded nanometre-sized 'cubosomes'. The authors suggested these 'cubosomes' might be expected to have interesting properties as novel drug delivery vehicles [224].

Another important development, which in the longer term should simplify the process of excipient selection, is the use of artificial neural networks to predict microemulsion phase behaviour [196].

5.9. Alternative surfactants

Another area of growing interest is the use of fluorinated surfactants for the stabilisation of microemulsion systems [225]. Fluorosurfactants are more surface-active than their hydrocarbon counterparts, the CMCs are typically two orders of magnitude lower, and importantly fluorosurfactants are less haemolytic [225]. The biomedical uses of fluorinated materials has been recently reviewed [226,227], and the low toxicity of a number of fluorinated solvents noted. Specific applications include blood substitutes such as Oxygent®, which is a low viscosity fluid emulsion comprising 60% fluorocarbon, perfluordecyl bromide as stabiliser, egg yolk lecithin as emulsifier and buffer [227]. Microemulsion formulations have also been described for use as a blood substitute, which employed a fluorocarbon oil and was stabilised by Montanox 80, a hydrogenated surfactant, which the authors claimed was a biocompatible [228,229]. Pulmonary aerosols based on fluorocarbons have also been indicated. The toxicological and regulatory status of fluorinated surfactants has not advanced to the point that they may be regarded as safe excipients in pharmaceutical formulations. Nevertheless, it is encouraging to observe the increasing number of reports describing the formation of microemulsion systems stabilised by fluorinated surfactants, or as surfactant mixtures with conventional hydrocarbon-based surfactants. In this context, it is also interesting to note that lipase and lipoxygenase have been successfully incorporated into a water-in-supercritical CO_2 microemulsions stabilised by a fluorosurfactant with retention of catalytic activity [230]. The incorporation of peptides and proteins into microemulsion systems stabilised by fluorosurfactants without loss of biological activity has therefore been clearly demonstrated. Fluorinated liposomes have been prepared using fluorocarbon–hydrocarbon diblocks [231,232], and the circulation time in mice of fluorinated phospholipid vesicles was prolonged three-fold in comparison to equivalent non-fluorinated vesicles [233]. The ability to use fluorinated amphiphiles in the formulation of emulsions, gels, microemulsions and vesicles indicate they have considerable potential as drug delivery systems, although the toxicological and regulatory hurdles will need to be overcome.

6. Conclusion

To date microemulsions have been shown to be able to protect labile drug, control drug release, increase drug solubility, increase bioavailability and reduce patient variability. Furthermore, it has proven possible to formulate preparations suitable for most routes of administration. There is still however a considerable amount of fundamental work characterising the physico-chemical behaviour of microemulsions that needs to be performed before they can live up to their potential as multipurpose drug delivery vehicles.

It is of interest to note that the first study investigating the use of microemulsions as potential drug delivery vehicles was reported in 1974 by Attwood et al. [234]. However the field lay virtually dormant until a review on the subject was published by Bhargava et al. in 1987 [43]. Since then there has been a very gradual increase in the number of research papers published on the topic until 1994 since when about 20 papers detailing the pharmaceutical use of microemulsions have been published each year. This small number of papers contrasts very sharply with liposomes where the number of publications dealing specifically with their use as drug delivery vehicles runs into the order of 300 per

year. Interestingly liposomes were first proposed as a delivery system in 1972, just a few years prior to Attwood and coworkers paper. This lack of research in the field does not mean that that microemulsions offer any less potential as delivery systems than liposomes, indeed it is pertinent to note that it took considerably less time for a microemulsion product (i.e. Neroal®) to get onto the market than the first liposomal drug delivery system.

References

[1] T.P. Hoar, J.H. Schulman, Transparent water-in-oil dispersions: the oleopathic hydro-micelle, Nature 152 (1943) 102–103.

[2] J.H. Schulman, W. Stoeckenius, L.M. Prince, Mechanism of formation and structure of micro emulsions by electron microscopy, J. Phys. Chem. 63 (1959) 1677–1680.

[3] I. Danielsson, B. Lindman, The definition of a microemulsion, Colloids and Surfaces 3 (1981) 391–392.

[4] K. Shinoda, B. Lindman, Organised surfactant systems: microemulsions, Langmuir 3 (1987) 135–149.

[5] L.M. Prince, A theory of aqueous emulsion. 1. Negative interfacial tension at the oil/water interface, J. Colloid Interface Sci. 23 (1967) 165–173.

[6] K. Shinoda, H. Kumieda, Conditions to produce so-called microemulsions. Factors to increase the mutual solubility of oil and water by solubilizer, J. Colloid Interface Sci. 42 (1973) 381–387.

[7] K. Shinoda, S. Friberg, Microemulsions. Colloidal aspects., Adv. Colloid Interface Sci. 4 (1975) 281–300.

[8] S. Friberg, I. Buraszcnska, Microemulsions in the water–potassium oleate–benzene system, Progr. Colloid Polymer Sci. 63 (1978) 1–9.

[9] E. Ruckenstein, J.C. Chi, Stability of microemulsions, J. Chem. Soc. Faraday Trans. 71 (1975) 1690–1707.

[10] J.Th.G. Overbeek, Microemulsions, a field at the border between lyophobic and lyophilic colloids, Faraday Disc. Chem. Soc. 65 (1978) 7–19.

[11] E. Ruckenstein, R. Krishnan, Effect of electrolytes and mixtures of surfactants on the oil–water interfacial tension and their role in formation of microemulsions, J. Colloid Interface Sci. 76 (1980) 201–211.

[12] T.F. Tadros, Microemulsions — an overview, in: K.L. Mittal, B. Lindman (Eds.), Proc. 4th Int. Symp. Surfactants in Solution, Plenum, New York, 1984, pp. 1501–1532.

[13] J. Eastoe, B.M.H. Cazelles, D.C. Steytler, J.D. Holmes, A.R. Pitt, T.J. Wear, R.K. Heenan, Water-in-CO_2 microemulsions studied by small-angle neutron scattering, Langmuir 13 (1997) 6980–6984.

[14] Y. Ikushima, N. Saito, K. Hatakeda, M. Arai, Water/AOT/ethane microemulsion under supercritical conditions as a reaction medium, J. Supercrit. Fluids 13 (1998) 217–224.

[15] R.M. Evans, S.J. Farr, The development of novel, pressurised aerosols formulated as solutions, J. Biopharm. Sci. 3 (1992) 33–40.

[16] N. Patel, M. Marlow, M.J. Lawrence, Microemulsions: a novel pMDI formulation, in: Drug Delivery to the Lungs IX, London, The Aerosol Society, Bristol, 1998, pp. 160–163.

[17] H. Sagitani, S. Friberg, Microemulsion systems with a non-ionic cosurfactant, J. Disper. Sci. Technol. 1 (1980) 151–164.

[18] D.W. Osborne, C.A. Middleton, R.L. Rogers, Alcohol-free microemulsions, J. Dispersion Sci. Technol. 9 (1988) 415–423.

[19] D. Attwood, C. Mallon, C.J. Taylor, Phase studies of oil-in-water phospholipid microemulsions, Int. J. Pharm. 84 (1992) R5–R8.

[20] R. Aboofazeli, N. Patel, M. Thomas, M.J. Lawrence, Investigations into the formation and characterisation of phospholipid microemulsions. IV. Pseudo-ternary phase diagrams of systems containing water–lecithin–alcohol and oil; the influence of oil, Int. J. Pharm. 125 (1995) 107–116.

[21] R. Aboofazeli, C.B. Lawrence, S.R. Wicks, M.J. Lawrence, Investigations into the formation and characterisation of phospholipid microemulsions. III. Pseudo-ternary phase diagrams of systems containing water–lecithin–isopropyl myristate and either an alkanoic acid, amine, alkanediol, polyethylene glycol alkyl ether or alcohol as cosurfactant, Int. J. Pharm. 111 (1994) 63–72.

[22] D. Attwood, A.T. Florence, Surfactant Systems: Their Chemistry, Pharmacy and Biology, Chapman and Hall, London, 1983.

[23] H.L. Rosano, J.L. Cavello, D.L. Chang, J.H. Whitham, Microemulsions: a commentary on their preparation, J. Soc. Cosmet. Chem. 39 (1988) 201–209.

[24] P.A. Winsor, Hydrotropy, solubilisation and related emulsification processes, J. Chem. Soc. Faraday Trans. 44 (1) (1948) 376–398.

[25] K. Shinoda, M. Araki, A. Sadaghiani, A. Khan, B. Lindman, Lecithin-based microemulsions: phase behaviour and microstructure, J. Phys. Chem. 95 (1991) 989–993.

[26] R. Aboofazeli, M.J. Lawrence, Investigations into the formation and characterization of phospholipid microemulsions: I Pseudo-ternary phase diagrams of systems containing water–lecithin–alcohol–isopropyl myristate, Int. J. Pharm. 93 (1993) 161–175.

[27] G.D. Rees, B.H. Robinson, Esterification reactions catalysed by *Chromobacterium viscosum* lipase in CTAB-based microemulsion systems, Biotechnol. Bioeng. 45 (1995) 344–355.

[28] M. Giustini, G. Palazzo, G. Colafemmina, M. Della Monica, M. Giomini, A. Ceglie, Microstructure and dynamics of the water-in-oil CTAB/*n*-pentanol/*n*-hexane/water microemulsion: spectroscopic and conductivity study, J. Phys. Chem. 100 (1996) 3190–3198.

[29] S.K. Mehta, X.X. Kawaljit, Isentropic compressibility and transport properties of CTAB-alkanol-hydrocarbon-water microemulsion systems, Colloids Surfaces A: Physicochem. Eng. Aspects 136 (1998) 35–41.

[30] I.S. Barnes, S.T. Hyde, B.W. Ninham, P.-J. Derian, M. Drifford, G.G. Warr, T.N. Zemb, The disordered open connected model of microemulsions, Progr. Colloid Polym. Sci. 76 (1988) 90–95.

[31] T. Skodvin, J. Sjöblem, J.O. Saeten, B. Gestblom, Solubilisation of drugs in microemulsions as studied by dielectric spectroscopy, J. Colloid Interface Sci. 155 (1993) 392–401.

[32] M. Olla, M. Monduzzi, L. Ambrosone, Microemulsions and emulsions in DDAB/water/oil systems, Colloids Surfaces A: Physicochem. Eng. Aspects 160 (1999) 23–36.

[33] K.A. Johnston, D.O. Shah, Effect of oil chain length and electrolytes on water solubilisation in alcohol-free pharmaceutical microemulsions, J. Colloid Interface Sci. 107 (1985) 269–271.

[34] M. Trotta, M.R. Gasco, F. Pattarino, Diffusion of steroid hormones from o/w microemulsions: influence of the cosurfactant, Acta Pharm. Technol. 36 (1990) 226–231.

[35] J. Bergenholtz, A.A. Romagnoli, N.J. Wagner, Viscosity, microstructure and interparticle potential of AOT/H2O/n-decane inverse microemulsions, Langmuir 11 (1995) 1559–1570.

[36] E. Acosta, D.H. Kurlat, M. Bisceglia, B. Ginzberg, L. Baikauskas, S.D. Romano, Induced electric birefringence and viscosity studies in microemulsions, Colloids Surfaces A: Physicochem. Eng. Aspects 106 (1996) 11–21.

[37] M. D'Angelo, D. Fioretto, G. Onori, L. Palmieri, A. Santucci, Dynamics of water-containing sodium bis(2-ethylhexyl)sulfosuccinate (AOT) reverse micelles: a high-frequency dielectric study, Phys. Rev. E 54 (1996) 993–996.

[38] J. Carlfors, I. Blute, V. Schmidt, Lidocaine in microemulsion — a dermal delivery system, J. Disp. Sci. Technol. 12 (1991) 467–482.

[39] J.N. Israelachvilli, D.J. Mitchell, B.W. Ninham, Theory of self assembly of hydrocarbon amphiphiles into micelles and bilayers, J. Chem. Soc. Faraday Trans. II 72 (1976) 1525–1567.

[40] D.J. Mitchell, B.W. Ninham, Micelles, vesicles and microemulsions, J. Chem. Soc. Faraday. Trans. II 77 (1981) 601–629.

[41] B. Jonsson, B. Lindman, K. Holmberg, B. Kronberg, Behaviour of concentrated surfactant systems, in: Surfactants and Polymers in Aqueous Solution, John Wiley & Sons, Chichester, 1999, pp. 61–89.

[42] W. Warisnoicharoen, A.B. Lansley, M.J. Lawrence, Nonionic oil-in-water microemulsions: the effect of oil type on phase behaviour, Int. J. Pharm. 198 (2000) 7–27.

[43] H.N. Bhargava, A. Narurkar, L.M. Lieb, Using microemulsions for drug delivery, Pharm. Tech. 11 (1987) 46–52.

[44] D. Attwood, Microemulsions, in: J. Kreuter (Ed.), Colloidal Drug Delivery Systems, Dekker, New York, 1994, pp. 31–71.

[45] J. Eccleston, Microemulsions, in: J. Swarbrick, J.C. Boylan (Eds.), Encyclopedia of Pharmaceutical Technology, Vol. 9, Marcel Dekker, New York, 1994, pp. 375–421.

[46] M.J. Lawrence, Surfactant systems: microemulsions and vesicles as vehicles for drug delivery, Eur. J. Drug Metab. Pharmacokinet. 3 (1994) 257–269.

[47] M.J. Lawrence, Microemulsions as drug delivery vehicles, Curr. Opin. Colloid Interface Sci. 1 (1996) 826–832.

[48] S. Tenjarla, Microemulsions: an overview and pharmaceutical applications, Crit. Rev. Therapeutic Drug Carrier Systems 16 (1999) 461–521.

[49] W. Warisnoicharoen, A.B. Lansley, M.J. Lawrence, Light scattering investigations on dilute non-ionic oil-in-water microemulsions, AAPS Pharm. Sci. 2 (2000) 429–448.

[50] F.C. Larche, P. Delord, Structures and stability of isotropic phases in the AOT–decane–water system, Fluid Phase Equilib. 20 (1985) 257–264.

[51] J. Eastoe, G. Fragneto, B.H. Robinson, T.F. Towey, R.K. Heenan, F.J. Lena, Variation of surfactant counterion and its effect on the structure and properties of Aerosol–OT-based water-in-oil microemulsions, J. Chem. Soc. Faraday Trans. 88 (1992) 461–467.

[52] P. Schurtenberger, Q. Peng, M.E. Leser, P.L. Luisi, Structure and phase behaviour of lecithin-based microemulsions: a study of chain length dependence, J. Colloid Interface Sci. 156 (1993) 43–51.

[53] P. Khoshnevis, S.A. Mortazavi, M.J. Lawrence, R. Aboofazeli, In-vitro release of sodium salicylate from water-in-oil phospholipid microemulsions, J. Pharm. Pharmacol. 49 (S4) (1997) 47.

[54] S.K. Mehta, R.K. Dewan, K. Bala, Percolation phenomenon and the study of conductivity, viscosity and ultrasonic velocity in microemulsions, Phys. Rev. E 50 (1994) 4759–4762.

[55] Z.J. Yu, R.D. Neuman, Reversed micellar solution-to-bicontinuous microemulsion transition in sodium bis(2-ethylhexyl) phosphate/*n*-heptane/water system, Langmuir 11 (1995) 1081–1086.

[56] R. Angelico, G. Palazzo, G. Colafemmina, P.A. Cirkel, M. Giustini, A. Ceglie, Water diffusion and head group mobility in polymer-like reverse micelles: evidence of a sphere-to-rod-to-sphere transition, J. Phys. Chem. B 102 (1998) 2883–2889.

[57] S.K. Mehta, X.X. Kavaljit, K. Bala, Phase behaviour, structural effects, volumetric and transport properties in nonaqueous microemulsions, Phys. Rev. E 59 (1999) 4317–4325.

[58] Y. Feldman, N. Kozlovich, I. Nir, N. Garti, Dielectric spectroscopy of microemulsions, Colloids Surfaces A: Physicochem. Eng. Aspects 128 (1997) 47–61.

[59] P.A. Cirkel, J.P.M. van der Ploeg, G.J.M. Koper, Branching and percolation in lecithin wormlike micelles studied by dielectric spectroscopy, Phys. Rev. E 57 (1998) 6875–6883.

[60] O. Regev, S. Ezrahi, A. Aserin, N. Garti, E. Wachtel, E.W. Kaler, A. Khan, Y. Talmon, A study of the microstructure of a four-component nonionic microemulsion by cryo-TEM, NMR, SAXS and SANS, Langmuir 12 (1996) 668–674.

[61] N. Nakamura, Y. Yamaguchi, B. Hakansson, U. Olsson, T. Tagawa, H. Kunieda, Formation of microemulsion and liquid crystal in biocompatible sucrose alkanoate systems, J. Disp. Sci. Technol. 20 (1999) 535–557.

[62] M. Kahlweit, R. Strey, D. Haase, H. Kunieda, T. Schmeling, B. Faulhaber, M. Borkovec, H.-F. Eicke, G. Busse, F.

Eggers, T.H. Funck, H. Richmann, L. Magid, O. Soderman, P. Stilbs, J. Winkler, A. Dittrich, W. Jahn, How to study microemulsions, J. Colloid Interface Sci. 118 (1987) 436–453.
[63] J. Alander, T. Warnheim, Model microemulsions containing vegetable oils Part 2: Ionic surfactant systems, JAOCS 66 (1989) 1661–1665.
[64] W.O. Parker Jr., C. Genova, G. Carignano, Study of micellar solutions and microemulsions of an alkyl oligoglucoside via NMR spectroscopy, Colloids Surfaces A: Physicochem. Eng. Aspects 72 (1993) 275–284.
[65] C. von Corswant, S. Engström, O. Söderman, Microemulsions based on soybean phosphatidylcholine and triglycerides. Phase behaviour and microstructure, Langmuir 13 (1997) 5061–5070.
[66] C. von Corswant, O. Soderman, Effect of adding isopropyl myristate to microemulsions based on soybean phosphatidylcholine and triglycerides, Langmuir 14 (1998) 3506–3511.
[67] C. von Corswant, C. Olsson, O. Soderman, Microemulsions based on soybean phosphatidylcholine and isopropyl myristate — effect of addition of hydrophilic surfactants, Langmuir 14 (1998) 6864–6870.
[68] C. von Corswant, P. Thoren, S. Engstrom, Triglyceride-based microemulsion from intravenous administration of sparingly soluble substances, J. Pharm. Sci. 87 (1998) 200–208.
[69] H. Saint Ruth, D. Attwood, G. Ktistis, C.J. Taylor, Phase studies and particle size analysis of oil-in-water phospholipid microemulsions, Int. J. Pharm. 116 (1995) 253–261.
[70] A. Shioi, M. Harada, M. Tanabe, Static light scattering from oil-rich microemulsions containing polydispersed cylindrical aggregates in sodium bis(2-ethylhexyl) phosphate system, J. Phys. Chem. 99 (1995) 4750–4756.
[71] G. Ktistis, Effect of polysorbate 80 and sorbitol concentration on in vitro release of indomethacin from microemulsions, J. Disp. Sci. Technol. 18 (1997) 49–61.
[72] C. Eugster, G. Rivara, G. Forni, S. Vai, Marigenol®-concentrates comprising Taxol and/or Taxan esters as active substances, Panminerva Medica 38 (1996) 234–242.
[73] P.P. Constantinides, J.P. Scalart, Formulation and physical characterisation of water-in-oil microemulsions containing long versus medium chain length glycerides, Int. J. Pharm. 158 (1997) 57–68.
[74] D. Hantzschel, S. Enders, H. Kahl, K. Quitzsch, Phase behaviour of quaternary systems containing carbohydrate surfactants–water–oil–cosurfactant, Phys. Chem. Chem. Phys. 1 (1999) 5703–5710.
[75] M.A. Bolzinger, M.A. Thevenin, J.L. Grossiord, M.C. Poelman, Characterisation of a sucrose ester microemulsion by freeze fracture electron micrograph and small angle neutron scattering experiments, Langmuir 15 (1999) 2307–2315.
[76] L. Auvray, J.P. Cotton, R. Ober, C. Taupin, Evidence for zero mean curvature microemulsions, J. Phys. Chem. 88 (1984) 4586–4589.
[77] J. Tabony, Formation of cubic structures in microemulsions containing equal volumes of water and oil, Nature 319 (1986) 400.
[78] J. Tabony, Occurrence of liquid-crystalline mesophases in microemulsion dispersions, Nature 320 (1986) 338–340.
[79] T.N. Zemb, I.S. Barnes, P.-J. Derian, B.W. Ninham, Scattering as a critical test of microemulsion structural models, Progr. Colloid Polym. Sci. 81 (1990) 20–29.
[80] M. Hirai, R.K. Hirai, H. Iwase, S. Arai, S. Mitsuya, T. Takeda, H. Seto, M. Nagao, Dynamics of w/o AOT microemulsions studied by neutron spin echo, J. Phys. Chem. Solids 60 (1999) 1359–1361.
[81] E.W. Kaler, S. Prager, A model of dynamic scattering by microemulsions, J. Colloid Interface Sci. 86 (1982) 359–369.
[82] J. Eastoe, K.J. Hetherington, D. Sharpe, J. Dong, R.K. Heenan, D. Steytler, Mixing of alkanes with surfactant monolayers in microemulsions, Langmuir 12 (1996) 3876–3880.
[83] T. Gulik-Krzywicki, K. Larsson, An electron microscopy study of the L2-phase (microemulsion) in a ternary system: triglyceride/monoglyceride/water, Chem. Phys. Lipids 35 (1984) 127–132.
[84] P.K. Vinson, J.G. Sheehan, W.G. Miller, L.E. Scriven, H.T. Davis, Viewing microemulsions with freeze-fracture transmission electron microscopy, J. Phys. Chem. 95 (1991) 2546–2550.
[85] M.A. Bolzinger, T.C. Carduner, M.C. Poelman, Bicontinuous sucrose ester microemulsion: a new vehicle for topical delivery of niflamic acid, Int. J. Pharm. 176 (1998) 39–45.
[86] D. Attwood, C. Mallon, G. Ktistis, C.J. Taylor, A study on factors influencing the droplet size in nonionic oil-in-water microemulsions, Int. J. Pharm. 88 (1992) 417–422.
[87] R. Aboofazeli, D.J. Barlow, M.J. Lawrence, Particle size analysis of concentrated phospholipid microemulsions. I. Total intensity light scattering, AAPS Pharm. Sci. 2 (2000) 449–470.
[88] J.M. Sarciaux, L. Acar, P.A. Sado, Using microemulsion formulations for drug delivery of therapeutic peptides, Int. J. Pharm. 120 (1995) 127–136.
[89] P.P. Constantinides, Lipid microemulsions for improving drug dissolution and oral absorption: physical and biopharmaceutical aspects, Pharm. Res. 12 (1995) 1561–1572.
[90] M. Trotta, S. Morel, M.R. Gasco, Effect of oil phase composition on the skin permeation of felodipine from (o/w) microemulsions, Pharmazie 52 (1997) 50–53.
[91] S. Kantaria, G.D. Rees, M.J. Lawrence, Gelatin-stabilised microemulsion-based organogels: rheology and application in iontophoretic transdermal drug delivery, J. Control. Rel. 60 (1999) 355–365.
[92] C. Malcolmson, C. Satra, S. Kantaria, A. Sidhu, M.J. Lawrence, Effect of oil on the level of solubilization of testosterone propionate into nonionic oil-in-water microemulsions, J. Pharm. Sci. 87 (1998) 109–116.
[93] V.H.L. Lee, Enzymatic barriers to peptide and protein absorption, Crit. Rev. Ther. Drug Carrier Syst. 5 (1988) 69–97.
[94] E.C. Swenson, W.J. Curatolo, Intestinal permeability enhancement for proteins, peptides and other polar drugs: mechanisms and potential toxicity, Adv. Drug Deliv. Rev. 8 (1992) 39–92.
[95] E.S. Swenson, W.B. Milisen, W. Curatolo, Intestinal per-

meability enhancement: efficacy, acute local toxicity and reversibility, Pharm. Res. 11 (1994) 1132–1142.
[96] P.P. Constantinides, J.-P. Scalart, S. Lancaster, J. Marcello, G. Marks, H. Ellens, P.L. Smith, Formulation and intestinal absorption enhancement evaluation of water-in-oil microemulsions incorporating medium-chain glycerides, Pharm. Res. 11 (1994) 1385–1390.
[97] P.P. Constantinides, C.M. Lancaster, J. Marcello, D.C. Chiossone, D. Omer, I. Hidalgo, P.L. Smith, A.B. Sarkahian, S.H. Yiv, A.J. Owen, Enhanced intestinal absorption of an RGD peptide from water-in-oil microemulsions of different composition and particle size, J. Control. Rel. 34 (1995) 109–116.
[98] C.W. Pouton, Formulation of self-emulsifying drug delivery systems, Adv. Drug. Del. Rev. 25 (1997) 47–58.
[99] M.R. Gasco, F. Pattarino, F. Lattanzi, Long-acting delivery systems for peptides: reduced plasma testosterone levels in male rats after a single injection, Int. J. Pharm. 62 (1990) 119–123.
[100] H. Willimann, P. Walde, P.L. Luisi, A. Gazzaniga, F. Stroppolo, Lecithin organogels as matrix for transdermal transport of drugs, J. Pharm. Sci. 81 (1992) 871–874.
[101] S. Murdan, B. van den Bergh, G. Gregoriadis, A.T. Florence, Water-in-sorbitan monostearate organogels (water-in-oil gels), J. Pharm. Sci. 88 (1999) 615–619.
[102] M. Kahlweit, G. Busse, B. Faulhaber, Preparing microemulsions with alkyl monoglucosides and the role of *n*-alkanols, Langmuir 11 (1995) 3382–3387.
[103] R. Duro, C. Souto, J.L. Gomez-Amoza, R. Martinez-Pacheco, A. Concheiro, Interfacial adsorption of polymers and surfactants: implications for the properties of disperse systems of pharmaceutical interest, Drug Devel. Ind. Pharm. 25 (1999) 817–829.
[104] A.G. Floyd, Top ten considerations in the development of parenteral emulsions, Pharm. Sci. Technol. Today 2 (1999) 134–143.
[105] Y.W. Cho, M. Flynn, Oral delivery of insulin, Lancet 2 (1989) 1518–1519.
[106] B. Fubini, M.R. Gasco, M. Gallarate, Microcalorimetric study of microemulsions as potential drug delivery systems. II. Evaluation of enthalpy in the presence of drugs, Int. J. Pharm. 50 (1989) 213–217.
[107] M. Gallarate, M.R. Gasco, M. Trotta, P. Chetoni, M.F. Saettone, Preparation and evaluation in vitro of solutions and o/w microemulsions containing levobunolol as ion pair, Int. J. Pharm. 100 (1993) 219–225.
[108] F. Pattarino, E. Marengo, M.R. Gasco, R. Carpignano, Experimental design and partial least squares in the study of complex mixtures: microemulsions as drug carriers, Int. J. Pharm. 91 (1993) 157–165.
[109] J.L. Murtha, H.Y. Ando, Synthesis of the cholesteryl prodrugs cholesteryl ibuprofen and cholesteryl flufenamate and their formulation into phospholipid microemulsions, J. Pharm. Sci. 83 (1994) 1222–1228.
[110] P. Schurtenberger, C. Cavaco, Polymer-like lecithin reverse micelles. 1. A light scattering study, Langmuir 10 (1994) 100–108.
[111] P.P. Constantinides, S.H. Yiv, Particle size determination of phase-inverted water-in-oil microemulsions under different dilution and storage conditions, Int. J. Pharm. 115 (1995) 225–234.
[112] K. Kriwet, C.C. Müller-Goymann, Diclofenac release from phospholipid drug systems and permeation through excised human stratum corneum, Int. J. Pharm. 125 (1995) 231–242.
[113] M.-J. Li, M.-H. Lee, C.-K. Shim, Inverse targeting of drugs to reticuloendothelial system-rich organs by lipid microemulsions emulsified by poloxamer 338, Int. J. Pharm. 113 (1995) 175–187.
[114] C. Satra, M. Thomas, M.J. Lawrence, The solubility of testosterone propionate in oil-in-water microemulsions, J. Pharm. Pharmacol. 47 (1995) 1126.
[115] M. Trotta, E. Ugazio, M.R. Gasco, Pseudo-ternary phase diagrams of lecithin-based microemulsions: influence of monoalkylphosphates, J. Pharm. Pharmacol. 47 (1995) 451–454.
[116] M.E. Leser, W.C. van Evert, W.G.M. Agterof, Phase behaviour of lecithin–water–alcohol–triacylglycerol mixtures, Colloids Surfaces A: Physicochem. Eng. Aspects 116 (1996) 293–308.
[117] M. Trotta, R. Cavalli, E. Ugazio, M.R. Gasco, Phase behaviour of microemulsion systems containing lecithin and lysolecithin as surfactants, Int. J. Pharm. 143 (1996) 67–73.
[118] A. Hasse, S. Keipert, Development and characterisation of microemulsions for ocular application, Eur. J. Pharm. Biopharm. 43 (1997) 179–183.
[119] M. Kahlweit, G. Busse, B. Faulhaber, Preparing nontoxic microemulsions. 2, Langmuir 13 (1997) 5249–5251.
[120] M. Trotta, F. Pattarino, G. Grosa, Formation of lecithin-based microemulsions containing *n*-alkanol phosphocholines, Int. J. Pharm. 174 (1998) 253–259.
[121] S.Y. Choi, S.G. Oh, S.Y. Bae, S.K. Moon, Effect of short-chain alcohols as cosurfactants on pseudo-ternary phase diagrams containing lecithin, Korean J. Chem. Eng. 16 (1999) 377–381.
[122] K.M. Park, M.K. Lee, K.J. Hwang, C.K. Kim, Phospholipid-based microemulsions of flurbiprofen by the spontaneous emulsification process, Int. J. Pharm. 183 (1999) 145–154.
[123] M. Trotta, Influence of phase transformation on indomethacin release from microemulsions, J. Control. Rel. 60 (1999) 399–405.
[124] M. Trotta, M. Gallarate, F. Pattarino, M.E. Carlotti, Investigation of the phase behaviour of systems containing lecithin and 2-acyl lysolecithin derivatives, Int. J. Pharm. 190 (1999) 83–89.
[125] C. von Corsant, P.E.G. Thoren, Solubilization of sparingly soluble active compounds in lecithin-based microemulsions: influence on phase behavior and microstructure, Langmuir 15 (1999) 3710–3717.
[126] A.H. Kibbe (Ed.), Handbook of Pharmaceutical Excipients, 3rd Edition, Pharmaceutical Press, London, 2000, pp. 407–423.
[127] C. Malcolmson, M.J. Lawrence, A comparison of the

incorporation of model steroids into non-ionic micellar and microemulsion systems, J. Pharm. Pharmacol. 45 (1993) 141–143.

[128] C. Malcolmson, M.J. Lawrence, Three-component non-ionic oil-in-water microemulsions using polyethylene ether surfactants, Colloids and Surfaces B: Biointerfaces 4 (1995) 97–109.

[129] J. Alander, T. Warnheim, Model microemulsions containing vegetable oils Part 1: Non-ionic surfactant systems, JAOCS 66 (1989) 1656–1660.

[130] N.J. Kale, L.V. Allen Jr., Studies on microemulsions using Brij 96 as surfactant and glycerin, ethylene glycol and propylene glycol as cosurfactants, Int. J. Pharm. 57 (1989) 87–93.

[131] H. Kunieda, N. Ushio, A. Nakano, M. Miura, Three-phase behaviour in a mixed sucrose alkanoate and poly-ethyleneglycol alkyl ether system, J. Colloid Interface Sci. 159 (1993) 37–44.

[132] W. Warisnoicharoen, A.H. Lansley, M.J. Lawrence, The cytotoxicity of nonionic oil-in-water microemulsions to a human cell line, in: Drug Delivery To the Lungs, Vol. VI, The Aerosol Society, 1995, pp. 40–43.

[133] K.-H. Oh, J.R. Baran, W.H. Wade, V. Weerasooriya, Temperature insensitive microemulsion phase behaviour with nonionic surfactants, J. Disper. Sci. Technol. 16 (1995) 165–188.

[134] S. Tolle, T. Zuberi, M.J. Lawrence, Physicochemical and solubilisation properties of *N,N*-dimethyl-*N*-(3-dodecyloxypropyl)-amineoxide: a biodegradable nonionic surfactant, J. Pharm. Sci. 89 (2000) 798–806.

[135] N.B. Desai, Esters of sucrose and glucose as cosmetic materials, Cosmetics and Toiletries 105 (2) (1990) 99–107.

[136] M. Kahlweit, G. Busse, B. Faulhaber, Preparing nontoxic microemulsions with alkyl monoglucosides and the role of alkanediols as cosolvents, Langmuir 12 (1996) 861–862.

[137] L.D. Ryan, E.W. Kaler, Role of oxygenated oils in *n*-alkyl β-D-monoglucoside microemulsions phase behaviour, Langmuir 13 (1997) 5222–5228.

[138] L.D. Ryan, K.-V. Schubert, E.W. Kaler, Phase behaviour of microemulsions made with *n*-alkyl monoglucosides and *n*-alkyl polyglycol ethers, Langmuir 13 (1997) 1510–1518.

[139] L.D. Ryan, E.W. Kaler, Effect of alkyl sulphates on the phase behaviour and microstructure of alkyl polyglucoside microemulsions, J. Phys. Chem. B 102 (1998) 7549–7556.

[140] C. Stubenrauch, B. Paeplow, G.H. Findenegg, Microemulsions supported by octyl monoglucoside and geraniol. 1. The role of alcohol in the interfacial layer, Langmuir 13 (1997) 3652–3658.

[141] S. Keipert, G. Schulz, Microemulsions with sucrose fatty ester surfactants. Part 1. In-vitro characterisation, Pharmazie 49 (1994) 195–197.

[142] M.A. Thevenin, J.L. Grossiord, M.C. Poelman, Sucrose esters/cosurfactant microemulsion systems for transdermal delivery: assessment of bicontinuous structures, Int. J. Pharm. 137 (1996) 177–186.

[143] N. Garti, V. Clement, M. Leser, A. Aserin, M. Fanun, Sucrose ester microemulsions, J. Molec. Liq. 80 (1999) 253–296.

[144] N. Garti, A. Aserin, M. Fanun, Non-ionic sucrose ester microemulsions for food applications. Part 1. Water solubilisation, Colloids Surfaces A: Physicochem. Eng. Aspects 164 (2000) 27–38.

[145] H.-O. Ho, C.C. Hsiao, M.-T. Sheu, Preparation of microemulsions using polyglycerol fatty acid esters as surfactant for the delivery of protein drugs, J. Pharm. Sci. 85 (1996) 138–143.

[146] P.P. Constantinides, G. Welzel, H. Ellens, P.L. Smith, S. Sturgis, S.H. Yiv, A.B. Owen, Water-in-oil microemulsions containing medium-chain fatty acids/salts: formulation and intestinal absorption enhancement evaluation, Pharm. Res. 13 (1996) 210–215.

[147] K.R. Wormuth, E.W. Kaler, Amines as microemulsion cosurfactants, J. Phys. Chem. 91 (1987) 611–617.

[148] R. Joubran, N. Parris, D. Lu, S. Trevino, Synergetic effect of sucrose and ethanol on formation of triglyceride microemulsions, J. Disp. Sci. Technol. 15 (1994) 687–704.

[149] M. Varshney, T. Khanna, M. Changez, Effects of AOT micellar systems on the transdermal permeation of glyceryl trinitrate, Colloids and Surface B: Biointerfaces 13 (1999) 1–11.

[150] M.J. Garcia-Celma, N. Azemar, M.A. Pes, C. Solans, Solubilisation of antifungal drugs in water/POE(20) sorbitan monooleate/oil systems, Int. J. Pharm. 105 (1994) 77–81.

[151] C. Nastruzzi, R. Gambari, Antitumor activity of (trans)dermally delivered aromatic tetra-amidines, J. Control. Rel. 29 (1994) 53–62.

[152] G. Kitstis, I. Niopas, A study on the in-vitro percutaneous absorption of propranolol from disperse systems, J. Pharm. Pharmacol. 50 (1998) 413–418.

[153] U. Schmalfuß, R. Neubert, W. Wohlrab, Modification of drug penetration into human skin using microemulsions, J. Control. Rel. 46 (1997) 279–285.

[154] M.E.A. Dalmora, A.G. Oliveira, Inclusion complex of piroxicam with beta-cyclodextrin and incorporation in hexadecyltrimethylammonium bromide based microemulsion, Int. J. Pharm. 184 (1999) 157–164.

[155] J. Kemken, A. Ziegler, B.W. Müller, Investigations into the pharmacodynamic effects of dermally administered microemulsions containing β-blockers, J. Pharm. Pharmacol. 43 (1991) 679–684.

[156] F. Dreher, P. Walde, P.L. Luisi, P. Elsner, Human skin irritation of a soybean lecithin microemulsion gel and of liposomes, Proc. Int. Symp. Control. Rel. Bioact. Mater. 22 (1995) 640–641.

[157] F. Dreher, P. Walde, P. Walther, E. Wehrli, Interaction of a lecithin microemulsion gel with human stratum corneum and its effect on transdermal transport, J. Control. Rel. 45 (1997) 131–140.

[158] W.A. Ritschel, Microemulsions for improved peptide absorption from the gastrointestinal tract, Meth. Find Exp. Clin. Pharmacol. 13 (1991) 205–220.

[159] K. Kriwet, C.C. Müller-Goyman, Mutual interactions between diclofenac diethylamine and phospholipids — investigations on the microstructure of the arisen systems, Pharmazie 49 (1994) 187–191.

[160] I. Siebenbrodt, S. Keipert, Poloxamer-systems as potential ophthalmics II. Microemulsions, Eur. J. Pharm. Biopharm. 39 (1993) 25–30.

[161] F. Pattarino, M.E. Carlotti, M.R. Gasco, Topical delivery systems for azelaic acid: effect of the suspended drug in microemulsion, Pharmazie 49 (1994) 72–73.

[162] M. Zabka, M. Benkova, Microemulsions containing local anaesthetics. Part 6: Influence of microemulsion vehicle on in vivo effect of pentacaine, Pharmazie 50 (1995) 703–704.

[163] M.R. Gasco, R. Cavalli, M.E. Carlotti, Timolol in lipospheres, Pharmazie 47 (1992) 119–121.

[164] S. Morel, M.R. Gasco, R. Cavalli, Incorporation in lipospheres of [D-Trp-6] LHRH, Int. J. Pharm. 105 (1994) R1–R3.

[165] R. Cavalli, D. Aquilano, M.E. Carlotti, M.R. Gasco, Study by X-ray powder diffraction and differential scanning calorimetry of two model drugs, phenothiazine and nifedipine, incorporated into lipid nanoparticles, Eur. J. Pharm. Biopharm. 41 (1995a) 329–333.

[166] R. Cavalli, S. Morel, M.R. Gasco, P. Chetoni, M.F. Saettone, Preparation and evaluation in vitro of colloidal lipospheres containing pilocarpine as ion pair, Int. J. Pharm. 117 (1995b) 243–246.

[167] R. Cavalli, O. Caputo, M.E. Carlotti, M. Trotta, C. Scarnecchia, M.R. Gasco, Sterilisation and freeze-drying of drug-free and drug-loaded solid lipid nanoparticles, Int. J. Pharm. 148 (1997) 47–54.

[168] R. Cavalli, O. Caputo, E. Marengo, F. Pattarino, M.R. Gasco, The effect of the components of microemulsions on both size and crystalline structure of solid lipid nanoparticles (SLN) containing a series of model molecules, Pharmazie 53 (1998) 392–396.

[169] S. Morel, E. Ugazio, R. Cavalli, M.R. Gasco, Thymopentin in solid lipid nanoparticles, Int. J. Pharm. 132 (1996) 259–261.

[170] R. Carpignano, M.R. Gasco, S. Morel, Optimisation of doxorubicine incorporation and of the yield of poly-butylcyanoacrylate nanoparticles, Pharm. Acta Helv. 66 (1991) 28–32.

[171] M.R. Gasco, S. Morel, M. Trotta, I. Viano, Doxorubicine englobed in polybutylcyanoacrylate nanocapsules: behaviour in vitro and in vivo, Pharm. Acta Helv. 66 (1991) 47–49.

[172] T.H. El-Faham, S.M. El-Shanawany, M.G. Abdel-Mohesen, In vitro and in vivo evaluation of a transparent oil–water gel formulation of flufenamic acid, Eur. J. Pharm. Biopharm. 38 (1992) 180–185.

[173] S. Bhatnagar, S.P. Vyas, Organogel-based system for transdermal delivery of propanolol, J. Microencapsulation 11 (1994) 431–438.

[174] F.P. Bonina, L. Montenegro, N. Scrofani, E. Esposito, R. Cortesi, E. Menegatti, C. Nastruzzi, Effects of phospholipid based formulations on in vitro and in vivo percutaneous absorption of methyl nicotinate, J. Control. Rel. 34 (1995) 53–63.

[175] F. Dreher, P. Walde, P.L. Luisi, P. Elsner, Human skin irritation studies of a lecithin microemulsion gel and of lecithin liposomes, Skin Pharmacol. 9 (1996) 124–129.

[176] R.W. Greiner, D.F. Evans, Spontaneous formation of a water-continuous emulsion from water-in-oil microemulsion, Langmuir 6 (1990) 1793–1796.

[177] N.H. Shah, M.T. Carvajal, C.I. Patel, M.H. Infeld, A.W. Malick, Self-emulsifying drug delivery systems with polyglycolyzed glycerides for improving in vitro dissolution and oral absorption of lipophilic drugs, Int. J. Pharm. 106 (1994) 15–23.

[178] S.A. Charman, W.N. Charman, M.C. Rogge, T.D. Wilson, F.J. Dutko, C.W. Pouton, Self-emulsifying drug delivery systems: formulation and biopharmaceutical evaluation of an investigational lipophilic compound, Pharm. Res. 9 (1992) 87–93.

[179] B. Matuszewska, L. Hettrick, J.V. Bondi, D.E. Storey, Comparative bioavailability of L-683,453, a 5α-reductase inhibitor, from a self-emulsifying drug delivery system in Beagle dogs, Int. J. Pharm. 136 (1996) 147–154.

[180] D.Q.M. Craig, S.A. Barker, D. Banning, S.W. Booth, An investigation into the mechanisms of self-emulsification using particle size analysis and low frequency dielectric spectroscopy, Int. J. Pharm. 114 (1995) 103–110.

[181] K. Hutchison, Digestible emulsions and microemulsions for optimum oral delivery of hydrophobic drugs, B.T. Gattefossé 87 (1994) 67–74.

[182] W.A. Ritschel, Microemulsions for improved peptide absorption from the gastrointestinal tract, Meth. Find. Exp. Clin. Pharmacol. 13 (1993) 205–220.

[183] W.A. Ritschel, Microemulsion technology in the reformulation of cyclosporine: the reason behind the pharmacokinetic properties of Neoral, Clin. Transplant. 10 (1996) 364–373.

[184] D.W. Holt, E.A. Mueller, J.M. Kovarik, J.B. van Bree, K. Kutz, The pharmacokinetics of Sandimmune Neoral: a new oral formulation of cyclosporine, Transplant. Proc. 26 (1994) 2935–2939.

[185] J.M. Kovarik, E.A. Mueller, J.B. van Bree, W. Tetzloff, K. Kutz, Reduced inter and intraindividual variability in cyclosporine pharmacokinetics from a microemulsion formulation, J. Pharm. Sci. 83 (1994) 444–446.

[186] E.A. Mueller, J.M. Kovarik, J.B. van Bree, W. Tetzloff, J. Grevel, K. Kutz, Improved dose linearity of cyclosporine pharmacokinetics from a microemulsion formulation, Pharm. Res. 11 (1994) 301–304.

[187] S. Noble, A. Markham, Cyclosporin: a review of the pharmacokinetic properties, clinical efficacy and tolerability of a microemulsion-based formulation (Neoral®), Drugs 50 (1995) 924–941.

[188] G.D. Rees, M.D.G. Nascimento, T.R.J. Jenta, B.H. Robinson, Reverse enzyme synthesis in microemulsion-based organogels, Biochim. Biophys. Acta 1073 (1991) 493–501.

[189] P.-Y. Yeh, P.L. Smith, H. Ellens, Effect of medium chain glycerides on the physiological properties of rabbit intestinal epithelium in vitro, Pharm. Res. 11 (1994) 1148–1154.

[190] R.A. Myers, V.J. Stella, Systemic bioavailability of penclomedine (NSC-338720) from oil-in-water emulsions administered intraduodenally in rats, Int. J. Pharm. 78 (1992) 217–226.

[191] T.T. Kararli, T.E. Needham, M. Griffin, G. Schoenhard,

L.J. Ferro, L. Alcom, Oral delivery of a renin inhibitor compound using emulsion formulations, Pharm. Res. 9 (1992) 888–893.

[192] J. Kemken, A. Ziegler, B.W. Müller, Influence of supersaturation on the pharmacodynamic effect of bupranol after dermal administration using microemulsions as a vehicle, Pharm. Res. 9 (1992) 554–558.

[193] M. Fujii, K. Shiozawa, T. Henmi, S. Yamanouchi, H. Suzuki, N. Yamashita, M. Matsumoto, Skin permeation of indomethacin from gel formed by fatty-acid ester and phospholipid, Int. J. Pharm. 137 (1996) 117–124.

[194] M.J. Garcia-Celma, Solubilisation of drugs in microemulsions, Indust. Appl. Microemulsions 66 (1997) 123–145.

[195] C. Satra, M. Thomas, M.J. Lawrence, Formulating oil-in-water microemulsions for pulmonary drug delivery, in: Drug Delivery To the Lungs, Vol. VI, The Aerosol Society, 1995, pp. 40–43.

[196] C.J. Richardson, A. Mbanefo, R. Aboofazeli, M.J. Lawrence, D.J. Barlow, Prediction of phase behaviour in microemulsion systems using artificial neural networks, J. Colloid Interface Sci. 187 (1997) 296–303.

[197] K. Kataoka, G.S. Kwon, M. Yokoyama, T. Okano, Y. Sakurai, Block copolymer micelles as vehicles for drug delivery, J. Control. Rel. 24 (1993) 119–132.

[198] X.C. Zhang, J.K. Jackson, H.M. Burt, Development of amphiphilic diblock copolymers as micellar carriers of taxol, Int. J. Pharm. 132 (1996) 195–206.

[199] X.C. Zhang, H.M. Burt, G. Mangold, D. Dexter, D. von Hoff, L. Mayer, W.L. Hunter, Anti-tumour efficacy and biodistribution of intravenous polymeric micellar paclitaxel, Anti-cancer Drugs 8 (1997) 696–701.

[200] T. Inoue, G. Chen, K. Nakamae, A.S. Hoffman, An AB block copolymer of oligo(methy methacrylate) and poly(acrylic acid) for micellar delivery of hydrophobic drugs, J. Control. Rel. 51 (1998) 221–229.

[201] B.G. Yu, T. Okano, K. Kataoka, G. Kwon, Polymeric micelles for drug delivery: solubilisation and haemolytic activity of amphotericin B, J. Control. Rel. 53 (1998) 131–136.

[202] J.E. Chung, M. Yokoyama, M. Yamato, T. Aoyagi, Y. Sakurai, T. Okano, Thermo-responsive drug delivery from polymeric micelles constructed using block copolymers of poly(*N*-isopropylacrylamide) and poly(butylmethacrylate), J. Control. Rel. 62 (1999) 115–127.

[203] Y. Matsumura, M. Yokoyama, K.Z. Kataoka, T. Okano, Y. Sakurai, T. Kawaguchi, T. Kakizoe, Reduction of the side effects of an anti-tumour agent, KRN5500, by incorporation of the drug into polymeric micelles, Jpn. J. Cancer Res. 90 (1999) 122–128.

[204] S.A. Hagan, A.G.A. Coombes, M.C. Garnett, S.E. Dunn, M.C. Davis, L. Illum, S.S. Davis, S.E. Harding, S. Purkiss, P.R. Gellert, Polylactide-poly(ethylene glycol) copolymers as drug delivery systems.1. Characterization of water dispersible micelle-forming systems, Langmuir 12 (1996) 2153–2161.

[205] P. Schmidt-Winkel, C.J. Glinka, G.D. Stucky, Microemulsion templates for mesoporous silica, Langmuir 16 (2000) 356–361.

[206] J.P. Spatz, S. Mossmer, M. Moller, Mineralisation of gold nanoparticles in a block copolymer microemulsion, Chemistry – A Eur. J. 2 (1996) 1552–1555.

[207] B. Michels, G. Waton, R. Zana, Dynamics of micelles of poly(ethylene oxide) poly(propylene oxide) poly(ethylene oxide) block copolymers in aqueous solutions, Langmuir 13 (1997) 3111–3118.

[208] R. Cortesi, E. Esposito, A. Maietti, E. Menegatti, C. Nastruzzi, Formulation study for the antitumor drug camptothecin: liposomes, micellar solutions and a microemulsion, Int. J. Pharm. 159 (1997) 95–103.

[209] R. Cortesi, C. Nastruzzi, Liposomes, micelles and microemulsions as new delivery systems for cytotoxic alkaloids, Pharm. Sci. Technol. Today 2 (1999) 288–298.

[210] P. Langguth, A. Kubis, G. Krumbiegel, W. Lang, H.P. Merkle, W. Wachter, L.H. Spahn, R. Weyhenmeyer, Intestinal absorption of the quaternary trospium chloride: permeability-lowering factors and bioavailabilities for oral dosage forms, Eur. J. Pharm. Biopharm. 43 (1997) 265–272.

[211] Z.-G. Gao, H.-G. Choi, H.-J. Shin, K.-M. Park, S.-J. Lim, K.-J. Hwang, C.-K. Kim, Physicochemical characterisation and evaluation of a microemulsion system for oral delivery of cyclosporin A, Int. J. Pharm. 161 (1998) 75–86.

[212] P. Erkko, H. Granlund, M. Nuutinen, S. Reitamo, Comparison of cyclosporin A pharmacokinetics of a new microemulsion formulation and standard oral preparation in patients with psoriasis, Brit. J. Derm. 136 (1997) 82–88.

[213] B. Hausen, T. Ikonen, N. Briffa, G.J. Berry, U. Christians, R.C. Robbins, L. Hook, N. Serkova, L.Z. Benet, W. Schuler, R.E. Morris, Combined immunosuppression with cyclosporine (Neoral) and SDZ RAD in non-human primate lung transplantation: systematic pharmacokinetic-based trials to improve efficacy and tolerability, Transplantation 69 (2000) 76–86.

[214] J.M. Kovarik, E.A. Muller, F. Richard, W. Tetzloff, Optimizing the absorption of valspodar, a P-glycoprotein modulator. Part II: Quantifying its pharmacokinetic variability and refining the bioavailability estimate, J. Clin. Pharmacol. 37 (1997) 1009–1014.

[215] E.A. Muller, J.M. Kovarik, Y. Uresin, S.S. Preisig-Fluckiger, S. Hensel, P.W. Lucker, B. Holt, Optimizing the absorption of valspodar, a P-glycoprotein modulator. Part I: Selecting an oral formulation and exploring its clinical pharmacokinetics/dynamics, J. Clin. Pharmacol. 37 (1997) 1001–1008.

[216] S.-M. Khoo, A.J. Humberstone, C.J.H. Porter, G.A. Edwards, W.N. Charman, Formulation design and bioavailability assessment of lipidic self-emulsifying formulations of halofantrine, Int. J. Pharm. 167 (1998) 155–164.

[217] K.M. Park, C.K. Kim, Preparation and evaluation of flurbiprofen-loaded microemulsions for parenteral delivery, Int. J. Pharm. 181 (1999) 173–179.

[218] M.B. Delgado-Charro, G. Iglesias-Vilas, J. Blanco-Méndez, M.A. López-Quintela, M.A.J.-P. Marty, R.H. Guy, Delivery of a hydrophilic solute through the skin from novel microemulsion systems, Eur. J. Pharm. Biopharm. 43 (1997) 37–42.

[219] H.O. Ho, M.C. Huang, L.C. Chen, A. Hsia, K.T. Chen, H.S. Chiang, B.W. Spur, P.Y.K. Wong, M.T. Sheu, The percutaneous delivery of prostaglandin E_1 and its alkyl esters by microemulsions, Chin. Pharm. J. 50 (1998) 257–266.
[220] S. Murdan, G. Gregoriadis, A.T. Florence, Novel sorbitan monostearate organogels, J. Pharm. Sci. 88 (1999) 608–614.
[221] S. Murdan, G. Gregoriadis, A.T. Florence, Non-ionic surfactant based organogels incorporating niosomes, S.T.P. Pharma Sci. 6 (1996) 44–48.
[222] C.C. Muller-Goymann, H.-J. Hamann, Sustained release from reverse micellar solutions by phase transformations into lamellar liquid crystals, J. Control. Rel. 23 (1993) 165–174.
[223] M. Scherlund, M. Malmsten, P. Holmqvist, A. Brodin, Thermosetting microemulsions as delivery systems for periodontal anesthesia, Int. J. Pharm. 194 (2000) 103–116.
[224] J. Gustafsson, H. Ljusberg-Wahren, M. Almgren, K. Larsson, Cubic lipid–water phase dispersed into submicron particles, Langmuir 12 (1996) 4611–4613.
[225] P. LoNostro, S.M. Choi, C.Y. Ku, S.H. Chen, Fluorinated microemulsions: a study of phase behaviour and structure, J. Phys. Chem. B 103 (1999) 5347–5352.
[226] M.P. Krafft, J.G. Riess, Highly fluorinated amphiphiles and colloidal systems, and their applications in the biomedical field. A contribution, Biochimie 80 (1998) 489–514.
[227] J.G. Riess, M.P. Krafft, Advanced fluorocarbon-based systems for oxygen and drug delivery, and diagnosis, Artificial Cells Blood Substitutes and Immobilisation Biotechnology 25 (1997) 43–52.
[228] C. Cecutti, I. Rico, A. Lattes, A. Novelli, A. Rico, G. Marion, A. Graciaa, J. Lachaise, New formulation of blood substitutes: optimisation of novel fluorinated microemulsions, Eur. J. Med. Chem. 24 (1989) 485–492.
[229] C. Cecutti, A. Novelli, I. Rico, A. Lattes, A new formulation for blood substitutes, J. Dispersion Sci. Technol. 11 (1990) 115–123.
[230] J.D. Holmes, D.C. Steytler, G.D. Rees, B.H. Robinson, Bioconversions in a water-in-CO_2 microemulsions, Langmuir 14 (1998) 6371–6376.
[231] J.G. Riess, Fluorinated vesicles, J. Drug Target. 2 (1994) 455–468.
[232] J.G. Riess, Introducing a new element — fluorine — into the liposomal membrane, J. Liposome Res. 5 (1995) 413–430.
[233] C. Santaella, F. Frezard, P. Vierling, J.G. Riess, Extended in vivo blood circulation time of fluorinated vesicles, FEBS Lett. 336 (1993) 481–484.
[234] D. Attwood, L.R.J. Currie, P.H. Elworthy, Studies of solubilized micellar solutions. I. Phase studies and particle size analysis of solutions with noionic surfactants, J. Colloid Interface Sci. 46 (1974) 249–256.
[235] D. Fennell Evans, H. Wennerstrom, Micro- and macroemulsions, in: The Colloidal Domain — Where Physics, Chemistry, Biology and Technology Meet. 1st Edition, Vol. 1, VCH Publishers, 1994, pp. 451–469.
[236] J. Israelachvili, The science and application of emulsions — an overview, Colloids Surf. A: Physicochem. Eng. Aspects 91 (1994) 1–8.
[237] P.J. Atkinson, B.H. Robinson, A.M. Howe, R.K. Heenan, Structure and stability of microemulsion-based organogels, J. Chem. Soc. Faraday Trans. 87 (1991) 3389–3397.